

(2) Copy of the declaration of co-inventors, Gary Van Nest, Gary Ott and Gail Barchfeld, dated May 14, 1997, submitted in related Application No. 08/418,870 ("Declaration-97");

(3) Copy of Ott et al., Vaccine Design: The Subunit and Adjuvant Approach, Chapter 10, Powell et al., Plenum Press, New York (Reference AH-2, filed with the supplemental information disclosure statement dated Jan 19, 1998);

(4) Statement of Grounds of Appeal to the Decision Revoking the European Patent, which was filed with the supplemental information disclosure statement dated Jan 19, 1998 (Reference AO-2);

(5) Copy of the declaration of Lynn Woodard (Document A-1 enclosed with the Statement of Grounds of Appeal to the Decision Revoking the European Patent); and

(6) Copy of Annex 2 to the Statement of Grounds of Appeal to the Decision Revoking the European Patent.

REMARKS

Introductory Comments:

Claims 1-5, 7-9, 29, 36, 38 and 39 were examined in the Office Action dated August 10, 1999 and rejected under 35 U.S.C. §103(a), as obvious. These rejections are traversed for the reasons discussed below. Applicants acknowledge and appreciate the withdrawal of the previous 35 U.S.C. §103(a) rejections over Hoskinson and Glass in view of Idson and Remington.

Rejections Under 35 U.S.C. § 103(a)

Claims 1-5, 7-9, 29, 36, 38 and 39 were rejected under 35 U.S.C. 103(a), as being unpatentable over Woodard et al., *Vaccine* 3:137-145 (1985) ("Woodard"), in view of Silvestri et al., *International Journal of Pharmaceutics*, 50:141-146 (1989) ("Silvestri") for reasons of record. The Office acknowledges that "neither Woodard nor Silvestri disclose an adjuvant composition consisting essentially of the exact range amounts of metabolizable oil and emulsifying agent in an oil-in-water emulsion as now claimed." Office Action, page 2. However, the Office concludes that it would have been obvious to

one of ordinary skill in the art to determine, as a matter of routine optimization, the amount of metabolizable oil and emulsifying agent required to make an adjuvant. Office Action, pages 2-3. Applicants respectfully traverse the rejection and disagree with the assertions made in support thereof for the following reasons.

As acknowledged by the Office, there is no suggestion in either of these references that formulations such as described in Woodard and Silvestri could be modified to render applicants' unique compositions. Additionally, there is absolutely no indication that doing so would be successful for producing an adjuvant composition as claimed.

It is well settled that *prima facie* obviousness can only be established if the following three basic criteria are met: (1) there must be some suggestion or motivation to modify the references, or to combine reference teachings; (2) there must be a reasonable expectation of success (for the modification and/or combination); and (3) the prior art reference(s) must teach or suggest all the claim limitations. M.P.E.P. §2143. Further, the fact that references can be combined or modified or that the claimed invention is well within the capabilities of one of ordinary skill in the art is not sufficient by itself to establish *prima facie* obviousness. *In re Mills*, 16 USPQ2d 1430 (Fed. Cir. 1990); *Ex parte Levengood*, 28 USPQ2d 1300 (BPAI 1993). Applicants respectfully submit that the Office has failed to establish *prima facie* obviousness, and that there is no suggestion to combine the teachings of the art as asserted. Applicants respectfully submit that the invention as a whole is not obvious and that there is no suggestion to combine the teachings of the art as asserted.

The claimed invention encompasses adjuvant compositions, formed from the combination of a metabolizable oil, wherein the oil is present in an amount of 0.5% to 20% of the total volume, and an emulsifying agent, wherein the emulsifying agent is 0.01% to 2.5% by weight (w/w), wherein the oil and said emulsifying agent are present in the form of an oil-in-water emulsion having submicron oil droplets. The claimed adjuvant composition exists in the absence of any polyoxypropylene-polyoxyethylene block copolymer and any muramyl peptide. Additionally, the claimed adjuvant composition is capable of increasing the immune response to an antigen when

administered with the antigen. In this regard, applicants direct the Office's attention to Declaration-94 (see paragraph 3), Declaration-97 (see paragraph 5), Examples 1-4 of the instant application, and Ott et al. (Reference AH-2), wherein the surprisingly superior adjuvant properties of the claimed adjuvant compositions have been clearly established. (A copy of each of the declarations and Ott et al. accompanies this response, for the Examiner's convenience).

Applicants reiterate that Woodard's compositions are distinct from the claimed invention. As is stated clearly in the title of the reference, Woodard's oil-in-water (O/W) emulsions are used as vehicles for adjuvants, rather than as adjuvants *per se*. In contrast, the O/W emulsions of the invention act as adjuvants themselves when the oil droplets are in the submicron range, rather than just as vehicles for adjuvants. Thus, in order to arrive at the currently claimed invention, a person skilled in the art would have to disregard the whole thrust of Woodard, namely that the emulsions are vehicles for adjuvants. Further, the adjuvant activity of the claimed emulsions is clearly in excess of any activity which might have been expected by the skilled person based on Woodard. In fact, in his affidavit Woodard concludes that "the observation that the emulsions with droplets less than 1 μ m are more effective adjuvants than emulsions equivalent in composition but larger droplets was an outstanding finding" (emphasis added). (See Woodard's affidavit, paragraph 46, accompanying this response for the Examiner's convenience).

Further, unlike the compositions disclosed in Woodard, the claimed adjuvant compositions do not contain an antigen. In particular, Woodard discloses water-in-oil (W/O) and oil-in-water (O/W) emulsions wherein the antigen is added either to the aqueous phase or the oil phase before mixing of the phases (emphasis added) (see page 139). Further, Woodard teaches that the antigen must be added to the internal phase (oil in O/W and water in W/O emulsions), for optimal antibody response (emphasis added). Woodard also teaches that addition of the antigen to the external (continuous) phase reduced antibody production considerably (emphasis added) (see page 142, right column). Thus, the emulsions disclosed by Woodard necessarily comprise an antigen.

By contrast, applicants' claimed adjuvant composition consists of O/W emulsions having oil droplets substantially all of which are less than 1 micron in diameter, wherein

the adjuvant composition is capable of increasing the immune response to an antigen when administered with the antigen. The antigen is co-administered with the preformed O/W emulsions adjuvant composition. As acknowledged by Woodard in his affidavit, based on the state of the art at the time of the invention, addition of the antigen to the preformed emulsion was contrary to the conventional wisdom that the antigen should be added to the oil or internal phase for maximal antibody response (see paragraphs 23 and 46 of Woodard's affidavit).

Additionally, applicants' adjuvant formulations have been shown to exhibit superior immunogenicity in the *in vivo* studies described in Examples 1-4, Declaration-94 (see paragraph 3), Declaration-97 (see paragraph 5), and by Ott et al. (Reference AH-2). In particular, in discussing the commercial embodiment of the invention known as "MF59TM adjuvant," Ott et al. state that "overall MF59 and related microfluidized low-oil emulsions generate antibody titres consistently higher than those obtained with aluminum hydroxide, equal to or higher than IFA, and equal to or lower than CFA/IFA." (See page 283 of the reference). These superior adjuvant properties are due to applicants' nonobvious adjuvant compositions containing emulsions with submicron droplets as claimed. In this regard, applicants' adjuvant compositions call for the formation of an oil-in-water emulsion consisting essentially of a metabolizable oil, wherein the oil is present in an amount of 0.5% to 20% of the total volume, and an emulsifying agent, wherein the emulsifying agent is 0.01% to 2.5% by weight (w/w), and wherein the emulsion has oil droplets substantially all of which are less than 1 micron in diameter, parameters that are neither taught nor suggested by the art cited against the claims.

The submicron size of the oil droplets in the emulsion in the claimed composition is demonstrated by the sizing data discussed in Declaration-94 (see paragraphs 4 and 5) and Ott et al. (Reference AH-2). The adjuvant composition was prepared according to the materials and methods disclosed in the application, and the size of the oil droplets was analyzed by laser light-scattering in the Malvern Mastersizer X using the lens system suitable for size determination in the 0.1 to 80 μ range. As illustrated in Figure 3 (Declaration-94) and Figure 10 (Ott et al.), the oil particles are submicron, with an average diameter of 0.36 μ . As discussed above, and as acknowledged by Woodard in his

affidavit, the submicron particle size rather than the composition is the major determinant of the adjuvant activity (see paragraph 21 of Dr. Woodard's affidavit).

Thus, as discussed above, and particularly as discussed in Dr. Woodard's affidavit (see paragraph 8), the methods described by Woodard would not be useful to make applicants' compositions as claimed. Further, there is no teaching or suggestion in Woodard to prepare an adjuvant composition as claimed, wherein the O/W emulsion contains submicron oil droplets, and further wherein the antigen is not added to the internal phase of the emulsion. Specifically, Woodard fails to teach or suggest adjuvant compositions that consist essentially of oil-in-water emulsion having oil droplets substantially all of which are less than 1 micron in diameter, formed from a metabolizable oil, wherein the oil is present in an amount of 0.5% to 20% of the total volume, and an emulsifying agent, wherein the emulsifying agent is 0.01% to 2.5% by weight (w/w), wherein the composition exists in the absence of any polyoxypropylene-polyoxyethylene block copolymer and any muramyl peptide, and further wherein the adjuvant composition is capable of increasing the immune response to an antigen when administered with the antigen. Thus, there is no motivation, deriving from Woodard to substantially modify the Woodard method in a way that would result in applicants' invention.

Additionally, as discussed in paragraph 5.17 of the Statement of Grounds of Appeal to the Decision Revoking the European Patent ("Appeal", Reference AO-2), none of the subsequently published papers that cite Woodard (Annex 2 to the Appeal) have developed Woodard's teaching towards the claimed invention. Most of these reference simply refer to the fact that Woodard had described an O/W emulsion, rather than disclosed the supposed adjuvant effect of O/W emulsions. Moreover, the references repeat the dogma that the antigen should be located within the oil droplets in an O/W emulsion for optimal antibody response, exemplifying that a skilled person would interpret Woodard as teaching away from the claimed invention (see paragraphs 6.11 to 6.15 of the Appeal). The teachings of Woodard are clearly in contrast to the submicron O/W emulsions of the current invention, which have been demonstrated to be effective. For example, "MF59TM adjuvant" has been tested in over 8000 human subjects, and is the only adjuvant other than alum to be licensed for human use, for example in Italy (see

paragraphs 6.16 to 6.17 of the Appeal). Thus, applicants submit that only in hindsight, and with the benefit of applicants' disclosure, could one of skill in the art arrive at the claimed invention.

Further, as acknowledged by the Office, Woodard does not teach the particularly claimed adjuvant compositions. Office action, page 2. Applicants submit that the Office has failed to consider the reference as a whole for what it fairly teaches to one of ordinary skill in the art. When the reference is considered in this manner, it is clear that Woodard does not teach or suggest applicants' specific claimed process.

Furthermore, as the Office correctly notes, none of the references taken individually teach the claimed adjuvant compositions. Office Action, page 2. However, the Office asserts that Silvestri provides the missing teaching. Applicants disagree.

Silvestri does not suggest immunological adjuvant compositions in the form of oil-in-water emulsion having droplets wherein substantially all of the droplets are less than 1 micron in diameter. In particular, Silvestri states that the submicron systems were investigated primarily because the submicron emulsions are more stable than equivalent emulsions (see page 142, right column). Further, the reference reiterates the teaching of Woodard, i.e., the submicron emulsions are used as drug delivery systems (see page 142, right column). There is no discussion regarding the use of submicron emulsions as adjuvants. Thus, the reference provides no suggestion or motivation to one skilled in the art to use the method of Silvestri to prepare the claimed adjuvant compositions. Additionally, the Office acknowledges that Silvestri does not disclose the claimed adjuvant composition. Office Action, page 2. Applicants therefore submit that Silvestri, when considered in its entirety, does not cure the deficiencies of Woodard.

Without the benefit of applicants' disclosure, the Office has failed to identify any motivation or suggestion to combine Woodard with Silvestri to arrive at the claimed adjuvant formulations. Such combination is therefore inappropriate. Thus, applicants submit that the claimed invention is nonobvious over the art and request reconsideration and withdrawal of this ground of rejection. However, if the rejection is not withdrawn, applicants request that the Examiner provide support for the above assertions by citing specific data in an affidavit pursuant to 37 C.F.R. §1.107(b). In the absence of such

objective evidence, applicants submit that the claimed invention is nonobvious over the art.

CONCLUSION

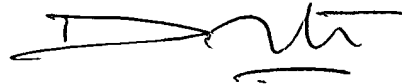
In view of the foregoing, applicants submit that the claims are now in condition for allowance and request early notification to that effect. Please direct all further communications regarding this application to:

Alisa A. Harbin, Esq.
CHIRON CORPORATION
Intellectual Property - R440
P.O. Box 8097
Emeryville, CA 94662-8097
Telephone: (510) 923-2708
Facsimile: (510) 655-3542.

Respectfully submitted,

Date: Nov 10, 1999

By: _____



Vandana Date
Registration No. 38,675
Attorney for Applicants

CHIRON CORPORATION
Intellectual Property - R440
P.O. Box 8097
Emeryville, CA 94662-8097
Telephone: (510) 923-2708
Facsimile: (510) 655-3542

CARPMAELS & RANSFORD

CHARTERED PATENT ATTORNEYS EUROPEAN PATENT ATTORNEYS TRADE MARK ATTORNEYS

43 BLOOMSBURY SQUARE
LONDON WC1A 2RA

AND AT MUNICH AND ALICANTE

TELEPHONE 0171 242 8692
TELEX 267209
FACSIMILE 0171 405 4166
0171 831 8501

email@london.carpmaels.com



IAN B P & M DEVAUX*
S DAVID VOTIER OBE*†
JOHN W M CARPMAEL*†
ALAN J JONES*
STEPHEN J COLGAN*
N KEITH HOWICK*†

ADRIAN J FISHER*†
CHRIS P MERCER*†
HUW G HALLYBONE*†
RICHARD E JACKSON*†
PAUL N HOWARD*†
ANNE WONG †

M J DONNAN*
P M JOHNSTON*
R W BISHOP†
A C W P JAMES*

R D HAWKINS *
PATRICIA HARRIS †
ANNA MEANLEY*†
SUSAN THOMAS *

JANDAN ALISS†

J A MURPHY (MANAGER)

CONSULTANT
DEREK G R GRUNDY

European Patent Office
Munich

YOUR REF T1071/97-332

OUR REF O00788EP/HGH/CJM

Dear Sirs

Re: European Patent 0399843
(ex 90305744.6)
Chiron Corporation

Further to the Notice of Appeal filed on 27th October 1997 in relation to the above-mentioned European patent, I hereby provide a statement setting out the grounds of appeal pursuant to Article 108 EPC.

I enclose, in triplicate, a new set of claims which constitutes the Appellant's First Auxiliary Request before the Board of Appeal. The Main Request remains the same as the Main Request before the Opposition Division.

I also enclose an affidavit by Dr Lynn Woodard (document A1 herein), co-author of the prior art paper on which the Opposition Division solely based its reasons for revocation of the Opposed Patent (D1).

1 Proceedings before the Opposition Division

1.1 Proceedings before the Opposition Division commenced in April 1995 with the filing of Notices of Opposition by the two opponents. A comprehensive response to the Notices was filed in March 1996 accompanied by a new Main Request. No further written argument or comment was filed by either opponent in the proceedings, this despite a specific request from the Opposition Division in the summons to Oral Proceedings to specify remaining objections in connection with the amended claims.

1.2 At the Oral Proceedings, the Opponents presented completely new arguments and attempted to introduce new material into the proceedings. Opponent OII indeed adopted a wholly different approach to lack of inventive step relying upon a different document altogether as the closest prior art.

1.3 The sufficiency of the specification was accepted by the Opposition Division in the written decision. It stated:

The teaching of the patent in suit is not that certain combinations of metabolizable oils and emulsifying agents provide the desirable effect of immunstimulating [sic] activity, but submicron emulsions exhibit said effect ... To prepare submicron emulsions is considered to be within the merit of the skilled person in the art. Furthermore, the patent in suit does disclose that the Microfluidizer-method, is an appropriate method to produce submicron emulsions (page 11, lines 1-7). Thus, it is considered that the skilled person in the art is not forced to undertake undue burden of experimentation in order to produce the oil droplets claimed.

1.4 The novelty of the claims was also accepted. The Opposition Division stated:

The feature of claim 1 that "at least 80% of the oil droplets are less than 0.5µm in diameter, is neither explicitly nor implicitly disclosed in any prior art document.

1.5 However, the Opposition Division went on to revoke the Opposed Patent, on the basis of a single document D1, for failing to meet the requirements of Article 56 EPC.

1.6 The Appellant submits, for reasons set out fully below, that the Opposition Division's decision on inventive step as expressed in the decision was incorrect in law and on the facts before it.

2 Claim requests

2.1 The Main Request before the Board of Appeal is identical to the Main Request considered by the Opposition Division, and claim 1 of this request is in the following terms:

An adjuvant composition, comprising:

(1) a metabolizable oil and

(2) an emulsifying agent,

wherein said oil and said emulsifying agent are present in the form of an oil-in-water emulsion having oil droplets characterized in that at least 80% by number of said oil droplets are less than 0.5µm in diameter and wherein said composition does not include a block copolymer.

2.2 As mentioned above, the novelty of these claims is not an issue under appeal, nor is the sufficiency of the specification in relation to these claims. Furthermore, there is no question of added subject-matter.

First Auxiliary Request

2.3 The First Auxiliary Request comprises process claims corresponding to the claims in the Main Request, and claim 1 is in the following terms:

A process for the production of a vaccine composition, comprising the steps of adding an immunostimulating amount of an antigen to an immunostimulating amount of an adjuvant formulation, said adjuvant formulation comprising:

- (1) a metabolizable oil and*
- (2) an emulsifying agent,*

wherein said oil and said emulsifying agent are present in the form of an oil-in-water emulsion having oil droplets characterized in that substantially all of said oil droplets are less than 1µm in diameter, and wherein said composition does not include a block copolymer, characterized in that, in the process, said antigen is added to said adjuvant formulation after the preparation of said adjuvant formulation.

2.4 The Board will appreciate that these claims introduce a significant feature, namely that in the adjuvant compositions of the invention the antigen has been added to the formulation after preparation of the adjuvant formulation, *ie.* to the water phase of the oil-in-water emulsion. This is a further significant distinction over the prior art.

3 Outline of grounds for appeal

The Appellant submits that:

- The inventors of the Opposed Patent made a significant discovery, namely that oil-in-water submicron emulsions make effective adjuvants in vaccine compositions.
- The Opposed Patent was revoked on the ground of obviousness in light of a single prior art reference, namely D1.
- The compositions of the Opposed Patent are, however, very different from those taught or suggested by D1, and D1 contains no suggestions which could prompt the person skilled in the art to arrive at the claimed invention. This is confirmed by the main author of D1.
- The finding that the claimed adjuvants were "obvious" is based upon an unacceptable hindsight analysis of the invention.

For this reason, the Appellant disputes the Opposition Division's conclusions *vis-à-vis* inventive step and it is requested that its decision be set aside and the patent be maintained.

4 The invention

The Opposed Patent is based upon the discovery that oil-in-water submicron emulsions make effective adjuvants. This is set out in the Opposed Patent in the following terms:

Surprisingly, it has been found that a satisfactory adjuvant formulation is provided by a composition comprising a metabolizable oil and an emulsifying

agent, wherein the oil and the emulsifying agent are present in the form of an oil-in-water emulsion having oil droplets substantially all of which are less than 1µm in diameter [page 3, lines 18-20].

5 Inventive step of the Main Request

5.1 The adjuvants of the Opposed Patent are very different from those disclosed in the prior art. They are surprisingly effective and are utilised in a different manner from those disclosed in, for instance, D1.

5.2 However, the Opposed Patent was revoked by the Opposition Division for supposedly failing to meet the requirements of Article 56 EPC in light of D1. The Opposition Division held that:

Document D1, does clearly disclose that the emulsions by themselves, have a significant intrinsic adjuvant activity (page 137, abstract, lines 19-22), which is verified in table 7 (page 142) ... Thus, the skilled person in the art would expect that the emulsions of the patent as characterized in claim 1 (i.e. at least 80% of the oil droplets are less than 5µm [sic] in diameter), would also exhibit the same significant intrinsic adjuvant activity as it is disclosed in D1. [page 12 of its written decision]

5.3 It is submitted that the legal reasoning behind this decision is incorrect.

5.4 The Opposition Division accepted that the emulsions disclosed in D1 do not destroy the novelty of the claimed compositions:

The feature of claim 1 that "at least 80% of the oil droplets are less than 0.5µm in diameter, is neither explicitly nor implicitly disclosed in any prior art document.

5.5 The Opposition Division has not indicated why the skilled person might be motivated to adapt the disclosure of D1 in order to produce a composition according to claim 1. The skilled person appears to have been endowed with the desire to develop the emulsions disclosed in D1 into the emulsions of the invention without reason or purpose.

5.6 It is therefore submitted that the Opposition Division was incorrect in revoking the patent in light of D1 because the skilled person could not and would not have arrived at the claimed compositions purely by considering D1.

5.7 Furthermore, D1 is concerned with the preparation of oil-in-water emulsions for use as vehicles for adjuvants, rather than for use as adjuvants *per se*. This is clearly stated in the title of D1, for instance. In contrast, the claimed invention is based on the finding that oil-in-water emulsions can act as adjuvants in their own right when the oil droplets are in the submicron range, rather than simply as vehicles for adjuvants.

5.8 In order to arrive at the presently claimed invention, therefore, it is submitted that the skilled person faced with D1 would have to disregard the whole thrust of D1, namely

that the emulsions are vehicles for adjuvants. The Opposed Patent cannot be obvious over D1, since the skilled person would have to abandon the teaching of D1 in order to arrive at the claimed invention.

5.9 The discussion of inventive step at the oral proceedings before the Opposition Division revolved around the statement in the abstract of D1 that:

hexadecane-in-water emulsions had significant intrinsic adjuvant activity.

5.10 The only data point in the whole of D1 which bears any resemblance to this statement can be found in Table 7. The fifth row of the table describes the results obtained with a control composition containing "No avridine in either injection" and gives an ELISA absorbance of 0.786. This result refers to a single experiment in which two hexadecane-in-water emulsions were administered, both without the addition of the adjuvant avridine.

5.11 This figure of 0.786 was discussed at length during the Opposition oral proceedings and, on the basis of this single data point, the Opposition Division held that the claimed invention was obvious. The Appellant makes three main observations in this respect.

5.12 Firstly, some of the figures given in Table 7 are accorded statistical significance, as shown by a superscript "a" (eg. "0.913^a" in the third row). This is not the case for the 0.786 figure, and so this data must be interpreted accordingly. It is the realistic and objective teaching of D1 which must be considered, rather than an unsupported assertion contained in a single sentence of the abstract.

5.13 Secondly, any supposed "significant intrinsic adjuvant activity" possessed by the control composition in D1 was not sufficient for that composition to be considered as suitable for use as a stand-alone adjuvant. Indeed, as discussed above, to interpret D1 in this way would be to ignore the whole thrust of D1. It is clear from D1 that the author had not discovered any significant intrinsic adjuvant effect, otherwise he would not have continued in his research into oil-in-water emulsions as carriers for lipophilic adjuvants such as avridine and hexadecylamine. This contrasts with the submicron oil-in-water emulsions of the present invention. For instance, document D13, which is a discussion of a commercial embodiment of the invention known as "MF59™ adjuvant", states that:

Overall, MF59 and related microfluidized low-oil emulsions generate antibody titres consistently higher than those obtained with aluminium hydroxide, equal to or higher than IFA, and equal to or lower than CFA/IFA.

5.14 The adjuvant activity of the claimed emulsions is clearly in excess of any activity which might have been expected by the skilled person based on D1. The claimed adjuvant compositions offer advantages beyond any "intrinsic adjuvant activity" noted by the authors of D1; indeed, the main author of D1 states in his affidavit [A1] that:

The observation that emulsions with droplets less than 1µm are more effective adjuvants than emulsions equivalent in composition but with larger droplets was an outstanding finding. [conclusion, paragraph 46]

5.15 Thirdly, D1 must be considered without relying upon the knowledge which we now have relating to the adjuvanticity of submicron oil-in-water emulsions. The Opposition Division's decision amounts to saying that the skilled person would have realised in May 1989 that the claimed compositions would act as good adjuvants in their own right, based solely upon the teaching of D1. It is submitted that this view is based upon a hindsight analysis of D1 which draws upon facts which were not known to the skilled person in May 1989.

5.16 Faced with the finding that submicron oil-in-water emulsions can act as effective adjuvants in their own right, the skilled person would not have believed that this was obvious in light of the data presented in D1. It is only with hindsight that the Opposition Division and the Opponents are able to say that the present inventors' findings were "expected". It appears that the Opposition Division failed to put itself objectively in the position of the skilled person at the priority date, but rather have attempted to recreate the invention from the prior art. This is a wholly wrong way to approach the determination of inventive step.

5.17 In this respect, the Appellant has located the 14 subsequently published papers which cite D1 using the Science Citation Index database. None of these later references indicate that anybody skilled in the art has actually developed the work of D1 in the manner which is alleged to have been "obvious", despite the presence of a supposed "significant intrinsic adjuvant activity".

5.18 The terms in which D1 is described in these papers are set out in Annex 2 to this statement for completeness.

5.19 None of these references could remotely be interpreted as showing that D1 had pointing the skilled person towards the claimed invention. Most of these citations simply refer to the fact that the authors of D1 had described an oil-in-water emulsion, rather than to the disclosure of a supposed adjuvant effect of oil-in-water emulsions. It is only with hindsight that the disclosure of D1 could be said to make the present invention obvious.

5.20 It is clear, therefore, that there was no indication whatsoever in the prior art that the claimed submicron oil-in-water emulsions would make effective adjuvants.

6 Inventive step of the First Auxiliary Request

6.1 Even if D1 is considered to have prompted the skilled person to produce the claimed adjuvant compositions, it is submitted that the invention as defined in the First Auxiliary Request meets the requirement of the EPC.

6.2 The First Auxiliary Request is based around processes for the production of vaccine compositions. D1 specifically teaches the skilled person not to produce vaccines in the claimed manner and so it is submitted that these claims must be considered to be inventive in light of D1.

6.3 The vaccine preparations administered in the Opposed Patent are prepared by mixing adjuvant with antigen, as described on page 11, lines 8-9:

Antigen was added to the adjuvant formulations above after preparation. The antigen and emulsion were mixed by shaking.

6.4 In other words, the antigen is added to the adjuvant after mixing the phases of the submicron oil-in-water emulsion.

6.5 This process contrasts with that used in D1:

W/O and O/W emulsions with BSA added to either the aqueous phase or to the oil phase before mixing of the phases were prepared and injected into mice. [page 139]

6.6 This distinction is extremely important, because, in contrast with the process used in the Opposed Patent, the process described in D1 results in antigen being concentrated within the oil phase of an oil-in-water emulsion — the final products are very different (eg. see claim 30 of First Auxiliary Request).

6.7 In D1, for instance, the antigen BSA was “ground in the oil phase”, resulting in an oil/antigen mixture, to which warmed saline was added for the formation of an emulsion. This grinding contrasts with the method used in the Opposed Patent, where antigen is added to the already-formed emulsion by “shaking”.

6.8 Because this grinding method results in the antigen being present in oil before formation of an emulsion, D1 contains no disclosure of submicron emulsions in the absence of adjuvant. This is reflected in claim 29 of the First Auxiliary Request.

6.9 The phase distribution of antigen in the claimed vaccine formulations compared with those of D1 is clearly very different, as discussed by Dr Woodard in his affidavit.

6.10 Furthermore, D1 could not be clearer in teaching that antigen should be added to the oil phase and, therefore, that the method of manufacture described in the Opposed Patent should not be used:

***Determination of emulsion phase to which antigen should be added ...**
Antibody responses were significantly higher ($p < 0.01$) when BSA was added to the aqueous phase of Freund's incomplete adjuvant and to the oil phase of a 5% hexadecane emulsion with hexadecylamine adjuvant (Table 5). [page 139, right hand column]*

Placing the BSA in the saline continuous phase resulted in antibody levels no greater than BSA in saline-emulsifier without oil. [page 141, right hand column]

With O/W or W/O emulsions, protein antigens must be added to the internal (disperse) phase — oil in the former and water in the latter — for optimal antibody response. Addition of the antigen to the external (continuous) phase reduced antibody production considerably. [page 142, right hand column]

6.11 This was also emphasised by Dr Woodard in his later work:

Our previous study had confirmed that antigen must be added to the oil phase for optimal antibody response. [document A2, page 223]

As a prelude to these experiments, we designed stable, metabolizable oil-in-water emulsions of hexadecane ... The only drawback is the need for dry or lyophilised antigen to be mixed in the oil phase of the emulsion. [document A1, page 224]

To be effective, antigens must be incorporated into the internal phase of oil emulsions, i.e., the water phase of W/O emulsions and the oil phase of O/W emulsions ... concentrated in the internal phase. [document A3, page 293]

6.12 Furthermore, in the papers listed in Annex 2 which cite D1, Boersma *et al.* (A13) repeat the dogma that antigen should be located within the oil droplets in an oil-in-water emulsion — a clear example of a skilled person interpreting D1 as teaching away from the claimed invention.

6.13 Thus any “significant intrinsic adjuvant activity” which might be disclosed in D1 can only be in respect of preparations in which the antigen is concentrated within the oil droplets of an oil-in-water emulsion.

6.14 In addition, document D8, which was originally relied upon by Opponent I to demonstrate common general knowledge, states that:

It is desirable to incorporate the drug into the innermost phase of the emulsion in order to successfully exploit the advantages of an emulsion dosage form. [page 34, right hand column]

6.15 The belief that antigen should not be present in the aqueous phase can still be found today. For example, as late as March 1997 a review was published stating that:

O/w emulsions result in excellent antigen presentation and moderate targeting-... It is important to incorporate immunogen into the oil phase [A4, page 251]

6.16 This is in clear contrast to the sub-micron oil-in-water emulsions of the present invention, which have been demonstrated to be effective. For example, using a commercial embodiment of the invention known as “MF59™ adjuvant” [eg. see D13], these formulations have been tested in over 8000 human subjects and have been licensed as an adjuvant for human use in Italy in an influenza vaccine (“Fluad™ vaccine”).

6.17 The following passages from two papers detailing the success of the claimed submicron oil-in-water emulsion adjuvants also serve to demonstrate that antigen is added to a pre-formed emulsion (“MF59™ adjuvant”), rather than being ground into the oil phase, as taught by D1:

Final vaccine formulations were generated by mixing the commercial vaccine 1:1 with adjuvant emulsion. Adjuvant/vaccine mixtures were injected intramuscularly within 6h of mixing. [A5, page 1558]

At the time of each immunization, HBV was mixed with MF59 adjuvant, and animals were immunized within 1h of vaccine preparation. [A6, page 1169]

6.18 The claimed invention is clearly, therefore, a departure from the teaching of the prior art. D1, for instance, teaches the skilled person to do the opposite of that which is described in the claims. Indeed Dr Woodard, the main author of D1, expresses in his affidavit his surprise that the claimed compositions are effective.

6.19 It is apparent that there were no indications whatsoever in the prior art which would lead the skilled person to the claimed invention.

6.20 The appellant regrets that it was not in a position to present these clear arguments to the Opposition Division but, unfortunately, the Opponents chose to rely on completely new arguments at oral proceedings, having ignored the Opposition Division's invitation to file final submissions beforehand. Before oral proceedings, for example, Opponent I's view of inventive step over D1 concerned the choice of metabolisable oil, and Opponent II had never argued that D1 should be considered as the closest prior art. Regardless of this ambush, the Appellant has now had a chance to consider these arguments and has been able to demonstrate that the claimed subject-matter is clearly inventive over the disclosure of D1.

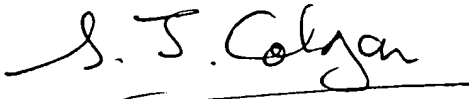
7 Requests

7.1 It is therefore requested that the Opposition Division's finding in relation to Article 56 EPC should be set aside and that the Opposed Patent be maintained with the claims of the Main Request filed herewith.

7.2 Alternatively, if the Board is minded to uphold the Opposition Division's decision, the Appellant requests that the Opposed Patent be maintained with the claims of the claims of the First Auxiliary Request or any further requests that may be admitted during the proceedings.

7.3 In the event that the Board is minded not to maintain the Opposed Patent, Oral Proceedings are requested.

Yours truly,



HALLYBONE, Huw George

(Authorised representative) **COLGAN, STEPHEN JAMES**

Annex 1 – Document list

The “D” documents are identical to those filed before the Opposition Division:

- D1 Woodard & Jasman (1987) Stable oil-in-water emulsions: preparation and use as vaccine vehicles for lipophilic adjuvants. *Vaccine* 3:137-144.
- D2 EP-A-0 382 271 (Akzo NV)
- D3 Yarkoni & Rapp (1980) Influence of type of oil and surfactant concentration on the efficacy of emulsified *Mycobacterium bovis* BCG cell walls to induce tumor regression in guinea pigs. *Infect Immun* 28:881-886.
- D4 Sanchez-Pescador *et al.* (1988) The effect of adjuvants on the efficacy of a recombinant herpes simplex virus glycoprotein vaccine. *J Immunol* 141:1720-1727.
- D5 Becher (1965) Emulsions: theory and practice, 2nd edition (Reinhold)
- D6 Kaufman & Garti (1981) Spectral absorption measurements for determination of ease of formation and stability of oil in water emulsions. *J Dispersion Sci Technol* 2:475-490.
- D7 Silvestri & Lostritto (1989) Theoretical evaluation of dispersed droplet radii in submicron oil-in-water emulsions. *Int J Pharmaceutics* 50:141-146.
- D8 Singh & Ravin (1986) Parenteral emulsions as drug carrier systems. *J Parenteral Sci Technol* 40:34-41.
- D9 EP-A-0 135 376 (Syntex USA Inc)
- D10 Herbert (1968) The mode of action of mineral-oil emulsion adjuvants on antibody production in mice. *Immunol* 14:301-318.
- D11 US patent 3,790,665
- D12 Intervet MasterSizer data
- D13 Ott *et al.* (1995) MF59, from *Vaccine design: the subunit and adjuvant approach*, eds. Powell & Newman (Plenum Press)
- D14 Affidavit of John D Barackman

The “A” documents are newly cited herein:

- A1 Affidavit of Lynn F Woodard
- A2 Woodard (1989) Adjuvant activity of water-insoluble surfactants. *Lab Anim Sci* 39:222-225.
- A3 Woodard (1990) Surface chemistry and classification of vaccine adjuvants and vehicles. From *Bacterial Vaccines*, pages 281-306.

- A4 Cox & Coulter (1997) Adjuvants — a classification and review of their modes of action. *Vaccine* 15:248-256.
- A5 Ott *et al.* (1995) Enhancement of humoral response against human influenza vaccine with the simple submicron oil/water emulsion adjuvant MF59. *Vaccine* 13:1557-1562.
- A6 Traquina *et al.* (1996) MF59 adjuvant enhances the antibody response to recombinant hepatitis B surface antigen vaccine in primates. *J Infect Dis* 174:1168-1175.
- A7 Warren & Chedid (1988) Future prospects for vaccine adjuvants. *CRC Crit Rev Immunol* 8:83-101.
- A8 Claasen *et al.* (1992) Freund's complete adjuvant: an effective but disagreeable formula. *Res Immunol* 143:478-483.
- A9 Lidgate *et al.* (1989) Formulation of vaccine adjuvant muramyl dipeptides. *Pharm Res* 6:748-152.
- A10 Holt *et al.* (1990) Immunisation of pigs with killed cultures of *Streptococcus suis* type 2. *Res Vet Sci* 48:23-27.
- A11 Ellis *et al.* (1991) Antigen specificity and activity of ovine antibodies induced by immunization with *Corynebacterium pseudotuberculosis* culture filtrate. *Vet Immunol Immunopathol* 28:303-316.
- A12 Kudrna *et al.* (1987) Immunologic memory responses induced in BALB/c mice by cross-linked outer membrane extracts of four *Salmonella* serotypes. *Am J Vet Res* 48:1199-1205.
- A13 Boersma *et al.* (1992) Adjuvant properties of the stable water-in-oil emulsions: evaluation of the experience with Specol. *Res Immunol* 143:503-512.
- A14 Azuma (1992) Synthetic immunoadjuvants: application to non-specific host stimulation and potentiation of vaccine immunogenicity. *Vaccine* 10:1000-1006.
- A15 Gupta *et al.* (1993) Adjuvants — a balance between toxicity and adjuvanticity. *Vaccine* 11:293-301.
- A16 Hilgers *et al.* (1994) A novel non-mineral oil based adjuvant. *Vaccine* 12:661-665.
- A17 Stieneker *et al.* (1995) Comparison of 24 different adjuvants for inactivated HIV-2 split whole virus as antigen in mice. *Vaccine* 13:45-53.
- A18 Todd *et al.* (1997) Development of an adjuvant-active nonionic block copolymer for use in oil-free subunit vaccines formulations. *Vaccine* 5:564-570.

Annex 2 – Later Published Papers Citing D1

Warren & Chedid [A7 herein]; page 86:

Different oils (such as peanut or sesame oils) and different emulsions (such as oil-in-water [cites D1] or water-in-oil-in-water) have been tried to optimize the adjuvant effect.

Claasen et al. [A8 herein]; page 479, right-hand column:

Water-soluble antigens can be emulsified with oil, or as double water-in-oil-in-water emulsions and lipophilic antigens as oil-in-water emulsions [cites D1].

Woodard [A2]; referring to his own previous work in D1:

page 222: *We have recently described stable O/W emulsions prepared with hexadecane as the hydrocarbon [cites D1].*

page 223: *Our previous study had confirmed that antigen must be added to the oil phase for optimal antibody response [cites D1].*

page 223: *Surfactant adjuvants and BSA were combined in the oil phase of an O/W emulsion as described [cites D1].*

page 224: *As a prelude to these experiments, we designed stable, metabolizable oil-in-water emulsions of hexadecane [cites D1].*

Lidgate et al. [A9 herein]; page 752:

Antibody response may not be contingent upon emulsion stability or globule size; biological response may occur as long as an emulsion is formed [cites D1].

Woodard [A3]; referring to his own previous work in D1:

page 286: *By combining the HLB system with spectrophotometric analysis of the emulsions, stable O/W emulsions suitable for vaccine and adjuvant carriers can be formulated for metabolizable n-alkanes and mineral oils [cites D1].*

page 290: *I have recently reexamined the structure activity of various aliphatic primary, secondary, tertiary, and quaternary aliphatic amine surfactants in an*

O/W emulsion [cites D1]. Except for avridine (discussed below), none of the aliphatic amines were any more adjuvant active than the emulsion alone.

page 293: *To be effective, antigens must be incorporated into ... the oil phase of O/W emulsions (discussed below) [cites D1].*

page 293: *Preparation of stable O/W emulsions with a 70:30 blend of Tween 80 and Span 80 as emulsifiers and hexadecane as the metabolizable oil has been recently described [cites D1].*

Holt et al. [A10 herein]; page 24, left-hand column:

To prepare the inoculation with FIA, formalin-killed, four-hour cultures containing 10^{11} organisms ml^{-1} , were emulsified with 5 per cent (v/v) Tween 80 and 20% (v/v) FIA [cites D1].

Ellis et al. [A11 herein]; page 305, in an apparent mistaken citation:

Equal amounts of filtrate and a block polymer adjuvant [cites D1] were emulsified.

Kudrna et al. [A12 herein]; page 1200, left-hand column, also an apparent mistake:

Preparation of vaccines — Vaccinal emulsions containing MDP were prepared, using emulsifiers and described procedures [cites D1].

Boersma et al. [A13 herein]:

page 504: *Emulsifying properties as well as stabilizing properties are required; other surfactants or combinations have also been applied [cites D1].*

page 505: *O/W emulsions, by their nature, are mostly used as vehicles with low viscosity for lipophilic compounds with adjuvant properties. [D1] also used Span as well as Tween compounds ... Best adjuvant properties were observed when the antigen was taken up in the oil fractions of the O/W. Addition of Tween to the water phase was detrimental to stability and adjuvant activity [cites D1].*

page 505: *Hexadecane is an effective oil phase whereas peanut oil, soybean oil or other plant-derived oils are far less effective [cites D1] ... The results with soybean oil could be improved by the addition of avridine [cites D1].*

page 505: *When O/W and W/O emulsions were compared, the W/O appeared to be most effective as an adjuvant [cites D1].*

Azuma [A14 herein]; page 1004:

Woodard and Jasman [cites D1] have also discussed stable o/w emulsion formulations.

Gupta et al. [A15 herein]; page 295:

Different emulsions such as oil-in-water [cites D1] and water-in-oil-in-water were also tried to get adjuvanticity.

Hilgers et al. [A16 herein]; page 665:

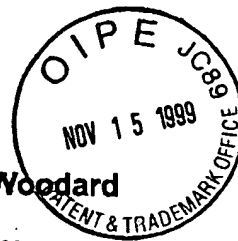
Additional active substances such as microbial glycolipids, synthetic block polymers of polyoxyethylene and polyoxypropylene with or without microbial products, avridine [cites D1] or SLP, can compensate for low activity of the biodegradable oil emulsions."

Stieneker et al. [A17 herein]; page 52, apparently mistakenly:

Toxic side-effects after application of FCA, FIA or Adjuvant 65 were not observed, although granuloma or subcutaneous abscesses have often been described by other authors [cites D1].

Todd et al. [A18 herein]; page 564:

Examples of adjuvants with carrier-mediated activity include aluminium and calcium-based salts, oil-based emulsions [cites D1] and liposomes.



A1

Affidavit of Lynn F Woodard

I, Lynn F Woodard, of PO Box 165, Tie Siding, Wyoming 82084, USA, hereby declare as follows:

1. I am the same LF Woodard who co-authored the paper entitled "Stable oil-in-water emulsions: preparation and use as vaccine vehicles for lipophilic adjuvants", published in 1985 in volume 3, pages 137-143, of the journal *Vaccine*, which I understand is referred to as document "D1" in these proceedings.
2. I received my BS (1968) and DVM (1970) degrees from Colorado State University and was self-employed as a veterinarian from 1970-1975. Graduate study at Washington State University resulted in a PhD degree in 1978. I was employed by the University of Idaho from 1978-1986 as an assistant and associate professor. In 1986, I took my current position as Head, Department of Veterinary Sciences, and Director, Wyoming State Veterinary Laboratory, at the University of Wyoming, USA.
3. I have read and understood a copy of European patent 0399843 ("the patent"), and I have been asked to comment on the patent, both in general and in the context of my own work as of May 1989.
4. I understand that the patent was revoked by the European Patent Office in July 1997 on the basis that it was obvious over my work as published in D1. For the reasons set out below, I do not agree with this conclusion.

Adjuvants

5. A "vaccine adjuvant" can be defined as a substance that is physically added with antigen(s) in an immunization to increase the immune response over that resulting from the administration of the antigen alone.
6. In May of 1989, aluminium salts (alums) and aluminium hydroxide gels were the only adjuvants approved for human use in the USA. Likewise, they were the vaccine adjuvants most used in veterinary vaccines. A few oil emulsion vaccines were in use in veterinary biologicals at that time. The composition and preparation of oil emulsion veterinary vaccines are not generally public information, but I believe that most are water-in-oil (W/O) or water-in-oil-in-water (W/O/W); most use a vegetable oil in the emulsion.
7. The laboratory "gold standard" of vaccine adjuvants is Freund's Complete Adjuvant (FCA or CFA). Developed by Jules Freund, it has been long used to stimulate both humoral (antibody-mediated) and cellular (cell-mediated) immunity. The composition of FCA is an approximate 50% water and 50% oil W/O emulsion with killed tuberculosis bacteria. Because of its very good ability to invoke antibody production and cell-mediated immunity, it is the standard by which adjuvants are typically judged. However, the composition is very tissue reactive and positive tuberculin skin tests result, so it is not appropriate for human or veterinary clinical use. The same preparation without tuberculosis bacteria is termed "Freund's Incomplete Adjuvant" (FIA or IFA). It is less reactive to tissue but still unacceptable for human use.

Concluded
11/28/99

8. My book chapter "Surface Chemistry and Classification of Vaccine Adjuvants" in the volume entitled "Bacterial Vaccines" (Advances in Biotechnological Processes, volume 13, Wiley-Liss, 1990), which I understand is referred to as A2 in these proceedings, gives a general review of adjuvants until 1990. Many additional review articles are cited in that chapter.
9. Although aluminum compounds are satisfactory adjuvants for many vaccines that afford protection by the production of antibodies, they generally do not stimulate cell-mediated immunity. The latter is generally needed to protect against many intracellular parasites such as viruses.

My research on oil-in-water emulsions

10. My PhD research examined the ability of newborn calves to mount cell-mediated immunity (CMI). The need to stimulate CMI led to a vaccine preparation developed by Edgar Ribi's group at NIH Rocky Mountain Laboratory. This vaccine contained an adjuvant component of the tuberculosis organisms found in FCA, namely trehalose dimycolate (TDM). This is a glycolipid that is very insoluble in water. The NIH group had found that TDM was most active when solubilized in oil droplets in an oil-in-water (O/W) emulsion, in contrast to the W/O emulsions developed by Freund.
11. Emulsions can be W/O or O/W depending on the type of emulsifiers used. A complete description of emulsions and the surfactants needed to prepare them can be found in, for instance, "Emulsions: Theory and Practice" by Paul Becher (2nd edition, Reinhold Publishing Co.), which I understand is document D5 in the present proceedings. Surface active agents, or "surfactants", are used to emulsify and stabilize the droplets. If the droplets are oil droplets suspended in water, an O/W emulsion is formed; if the droplets are water droplets suspended in oil, a W/O emulsion is formed. The droplets are termed the internal or disperse phase and the balance is termed the external, bulk or continuous phase.
12. In 1983, I set about developing O/W emulsions with the following characteristics: metabolizable, non-toxic, stable over time and temperature, and easy to prepare. O/W emulsions with which I had previously worked, and which were available in my laboratory, were not very stable.
13. Because W/O emulsions such as FIA cause severe chronic infection site reactions known as granulomas, a low oil concentration was desirable. Granulomas are totally unacceptable in humans and less than desirable in livestock and pets. In food animals, injection site lesions result in tough fibrotic areas that are unpalatable and that must therefore be trimmed, resulting in waste.
14. In a series of experiments, described in D1, we examined a variety of parameters that might affect emulsion stability and antibody response. In general, a 70:30 blend of Tween 80 and Span 80 emulsifiers ("T80/S80") mixed with an equal amount of hexadecane oil resulted in the most satisfactory emulsions. Equal 5% concentrations of oil and the T80/S80 blend were selected for subsequent experiments. Addition of bovine serum albumin (BSA) antigen into the internal (oil) phase tended to destabilize the emulsions and the addition of hexadecylamine adjuvant tended to stabilize the emulsion. Other Tween and Span emulsifiers were tested and three additional blends were found to be stable; one with T20/S80 was later shown to induce less antibody than T80/S80. An unstable micellar preparation with T21/S85 was also less effective.

15. Using BSA antigen, antibody titers were higher when the antigen was added to the internal phase of both O/W and W/O emulsions than to the external phase, at the 99% confidence interval ($p < 0.01$), as shown in Table 5 of D1. This was consistent with the understanding of FIA.
16. The protein would not be expected to "dissolve" in the oil, but it would be expected to be suspended in the oil. Over time, the proteins would be expected to move towards the aqueous phase with the hydrophobic amino acid sequences aligned along the water-oil interface.
17. The mechanism by which physically grinding the BSA into the oil of a O/W emulsion results in increased antibody was not understood. However, it confirmed previous work by the NIH Rocky Mountain Laboratory group that the antigen should be in the internal phase for optimal response. The NIH group had found this to be true for tumor regression and delayed hypersensitivity responses. Thus, our work confirmed previous findings that the antigen should be added to the internal phase of O/W or W/O emulsions for a variety of immune responses.
18. Keeping the 70:30 T80/S80 blend at a constant 5% while changing the oil concentration from 1% to 20% had no effect on antibody production with one exception. Likewise, keeping the oil at a constant 5% while reducing the T80/S80 to only 2% did not reduce antibody production. In general, stability of the emulsions did not affect antibody production to the BSA antigen.
19. In the final experiment, avridine adjuvant was used instead of the less potent hexadecylamine in the O/W emulsions. It was able to stimulate antibody titers equal to those induced by FIA. Increasing the oil and T80/S80 concentrations to 20% to increase surface area for adjuvant adhesion did not increase antibody responses, even when adjuvant concentration was increased 10-fold.
20. I continued researching oil-in-water emulsions after D1, and in 1989 a research article of mine, which I understand is referred to as A1 in these proceedings, was published in the journal *Laboratory Animal Science*. This developed my earlier work and compared the efficacy of several known adjuvants. I concluded this paper by stating that "our results show that adhesive spreading agent surfactants are satisfactory replacements for Freund's adjuvants when antigen and adjuvant are incorporated into the oil phase of dilute hexadecane emulsions."

European Patent 0399843

21. In my opinion, European Patent 0399843 refers to O/W emulsions with adjuvant activity when the oil droplets are submicron in size and made without the use of block copolymer emulsifiers. The invention is said to be that the emulsifiers need not have adjuvant activity themselves because size alone is sufficient for adjuvant activity. In other words, oil droplet size is the major determinant of adjuvant activity, rather than composition.
22. In the majority of examples, except as noted below, the submicron oil droplets were prepared by physically mixing squalene and muramyl tripeptide-phospholipid with or without T80 emulsifier and then passing the mixture through a Microfluidizer, resulting in 100 to 750nm oil droplet emulsions.
23. After formation of these emulsions, the antigen was added to the prepared emulsion and mixed by shaking. Addition after the emulsion was formed would mean that the antigen was added to the

external or bulk phase of the emulsion. Based on previous research, this is contrary to the notion that the antigen should be added to the oil or internal phase for maximal antibody response.

24. To assist in understanding the data contained in the patent, the following tables abstract comparative data from the results in the patent:

25. Goat immunization with HSV gD2 antigen

26. I have been asked to consider data in the patent relating to the effect of size on adjuvanticity. A number of different adjuvants were tested with different droplet sizes. These are designated MTP-PE-LO (10µm), MTP-PE-LO-KE (1-2µm), MF#13 (0.8µm) and MF#16 (0.5-0.6µm) These data are collated in the following table:

Comparison of: <ul style="list-style-type: none"> Table 6, groups 3 & 4 (page 21) Table 9, group 2 (page 26) Table 10 (page 27) All animals given 100µg HSV gD2 antigen, with a variety of adjuvants, using the same immunization protocol				
Adjuvants: <ul style="list-style-type: none"> MTP-PE-LO: 4% squalene, 100µg/ml MTP-PE, 0.008% Tween, ~10µm droplet size MTP-PE-LO-KE: 4% squalene, 100µg/ml MTP-PE, 0.008% Tween, ~1-2µm droplet size MF#13: 4% squalene, 100µg/ml MTP-PE, no Tween, microfluidized to ~0.8µm droplet size MF#16: 4% squalene, 500µg/ml MTP-PE, no Tween, microfluidized to 0.5-0.6µm droplet size 				
Data from	Animal #	Adjuvant	ELISA titers after	
			2nd immunization	3rd immunization
Table 6 (group 3)	3612	MTP-PE-LO	77	194
	3613	MTP-PE-LO	145	323
Table 6 (group 4)	3614	MTP-PE-LO-KE	123	227
	3615	MTP-PE-LO-KE	56	46
Table 9 (group 2)	4598	MF #13	2966	2207
	4599	MF #13	1661	Died
Table 10	5013	MF #16	1299	386
	5014	MF #16	6657	2806
	5015	MF #16	8206	1943
	5016	MF #16	7886	1514

27. Whilst these results are not ideal (eg. small sample size, no indication that age and breed of goat were not variables, variation in the control values), only one of the four goats immunized with the syringe-prepared (MTP-PE-LO) or Kirkland emulsified (MTP-PE-LO-KE) vaccines had a titre

exceeding the two CFA/IFA controls with the lowest titres. When the microfluidized vaccines (MF#13 & MF#16) were given, all 6 had titers with values that exceeded about 1300 after two doses. There does appear to be a real trend toward higher titers with the microfluidised MF preparations.

28. Comparing those goats that received a constant (100µg) amount of MTP-PE (groups 3 & 4 in Table 6 vs. group 2 in Table 9), the effect of emulsion preparation and droplet size can be studied. A trend of about a 10-fold increase in antibody can be seen when the droplet size is reduced to the submicron level. Again, the small sample size precludes any statistical evaluation, but as droplet size decreased to less than 1µm, there is a trend towards higher titers.
29. The presence of 0.008% T80 in MTP-PE-LO but not MF#13 preparations does not alter my interpretation of the results. This is too little T80 to have significant emulsifying power and no positive or negative effects on antigenicity would be expected.
30. Increasing the MTP-PE from 100µg (MF#13) to 500µg (MF#16) in microfluidized emulsions also gives increased titers. This increase could be due either to the increased amount of MTP and/or the decreased droplet size from ~0.8µm to 0.5-0.6µm.
31. Goat immunization with HIV gp120 antigen
32. I have been asked to consider data in the patent relating to the effect of MTP on adjuvanticity. The data in Table 11 of the patent are informative in this respect:

Comparison of: <ul style="list-style-type: none">• Table 11, groups 1, 3, 5 & 6 (pages 28-29) All animals given 100µg HIV gp120 antigen, with a variety of adjuvants, using the same immunization protocol			
Adjuvants: <ul style="list-style-type: none">• CFA/IFA: CFA for first immunization, IFA for second & third• MF#14: 4% squalene, 500µg/ml MTP-PE, no Tween• MF#15: 4% squalene, 100µg/ml MTP-PE, 0.008% Tween• MTP-PE-LO-KE: 4% squalene, 100mg/ml MTP-PE, 0.008% Tween			
Group	Adjuvant	ELISA titers after	
		2nd immunization	3rd immunization
1	CFA/IFA	1861±539	6630±996
3	MF #14	101±1089	1324±997
6	MF #15	10±333	3277±767
5	MTP-PE-LO-KE	721±416	632±32

33. These results would suggest that the CFA/IFA vaccine is superior after the second injection or third injection. Statistical evaluation of these results might be possible, but the large standard errors would make statistical differences difficult to detect with the small numbers involved.

34. Increasing the MTP-PE from 100µg (MF#15) to 500µg (MF#14) gave lower titers after the third immunization (*cf.* paragraph 30 above). Reducing the droplet size whilst maintaining the MTP-PE concentration at 100µg (MF#15 vs. MTP-PE-LO-KE) resulted in much higher titers after the third immunization. The method of emulsion preparation would appear to be responsible for this increased response.
35. MF#15 & MTP-PE-LO-KE are identical in relation to T80 and, as noted above, the T80 difference between groups 3 (MF#14) and 6 (MF#15) should not be of concern.
36. I conclude from these limited data that a dose-related response to MTP is not apparent. Also, the concentration of T80 does not seem to have consistent effect on adjuvanticity.
37. Goat immunization with HIV env antigen
38. A comparison of further data from Table 11 with that in Table 2 is also useful in considering the contribution of MTP towards adjuvanticity. In addition, these data demonstrate that submicron emulsions could be more effective than Freund's adjuvants:

Comparison of:			
<ul style="list-style-type: none"> Table 11, groups 2 & 4 (page 28): 100µg antigen Table 2 (page 14): 100µg antigen or more (up to 1mg) 			
All animals given HIV env antigen, with a variety of adjuvants, using the same immunization protocol			
Adjuvants:			
<ul style="list-style-type: none"> CFA/IFA: CFA for first immunization, IFA for second & third MF#14: 4% squalene, 500µg/ml MTP-PE, no Tween MTP-PE-LO: 4% squalene, 0-500µg/ml MTP-PE, 0.008% Tween, ~10µm droplet size 			
Group	Adjuvant	ELISA titers after	
		2nd immunization	3rd immunization
2	CFA/IFA	2235±680	5074±1378
4	MF #14	2351±1688	9896±2493
Table 2	MTP-PE-LO	typically <100	no data

39. Comparing groups 2 and 4 in Table 11 shows that the MF#14 preparation is very similar in eliciting antibodies to this antigen when compared to CFA/IFA after two doses. After 3 injections, responses were greater than with the "gold standard" Freund's. Even with the limited numbers, the responses may be near statistical significance although I have not performed any such calculations.
40. These titers are favorable when compared to the near total lack of response when the antigen was given with 20µg to 500µg of MTP-PE (Table 2) with a droplet size of ~10µm. Assuming that titres in Tables 2 & 11 were determined in the same manner, the method of emulsion preparation would appear to be the main determinant of increased adjuvanticity.

41. Again, the absence of T80 prevents a strict comparison, but its inclusion is not of concern, and it certainly had no positive effect.

42. Baboon immunization with HIV env antigen

43. I have been asked to consider the data in the patent which compare the submicron microfluidized adjuvants with other conventional adjuvants. One such comparison is given above in paragraph 39, but the data on pages 31-32 of the patent is also informative. I note that this data is presented in a table labeled as "Table 11" when it is, in fact, the twelfth table in the patent.

Comparison of:			
<ul style="list-style-type: none"> Table 12, groups 2, 4 & 6 (page 31) — misnumbered as Table 11 			
All animals given 25µg HIV env antigen, with a variety of adjuvants, using the same immunization protocol			
Adjuvants:			
<ul style="list-style-type: none"> MTP-PE/IFA: 250mg MTP-PE in IFA MF#1: 2% squalene, 500µg/ml MTP-PE, no Tween, 0.17µm droplet size Alum: 0.8mg/ml aluminum hydroxide 			
Group	Adjuvant	ELISA titer	Neutralizing antibody
2	MTP-PE/IFA	400	<10
		34500	30
		142300	200
4	MF #1	600	<10
		14400	35
		87400	>250
6	Alum	4900	80
		700	<10

44. This data shows that ELISA and neutralizing antibody responses were similar for IFA, MF#1 and alum preparations with the limited animals represented, although there was great variability in all groups. The same can be said for the baboon immunization using gp120, also shown on page 31.

Conclusions

45. Collectively, the data in the patent would support the following generalizations, realizing that statistical conclusions are not possible with the limited numbers tested in any given experiment. However, the following trends are noted:

- the method of mechanical emulsification does appear to affect antibody response: as more energy is added to the system of emulsification, leading to smaller droplet size, responses increase. Thus, MF preparations were superior to KE or syringe-prepared emulsions.
- the species of animal tested may greatly affect the response to a given antigen (eg. guinea pigs vs. goats)

- when using MTP-PE as both exogenous adjuvant and emulsifier, a dose-related response is not evident in higher mammals.
- size of droplets appears to be inversely related to antibody response with most antigens tested in higher mammals
- use of a very small amount of T80 did not appear to affect antibody response

46. When compared to the knowledge of the date of application (May 1989), I would state the following about the patent:

- the patent shows that droplet size of O/W emulsions is important. My work had shown that unstable emulsions with large droplets were just as effective with BSA antigen as stable emulsions with smaller droplets. The observation that emulsions with droplets less than 1µm are more effective adjuvants than emulsions equivalent in composition but with larger droplets was an outstanding finding.
- inclusion of the antigen in the aqueous phase rather than the internal oil phase of the O/W emulsion is contrary to my findings and recommendations for maximal antibody response.

Further affiant sayeth not.

Signed

Lynn F. Woodard

Lynn F Woodard

Date

12/29/97

Place

Laramie, WY, USA

Before me

Denise A. Baker

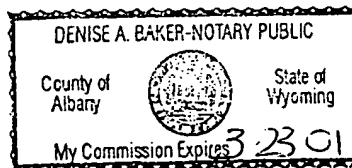
Notary Public

Date

12-29-97

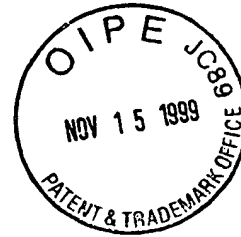
Place

Laramie WY



CURRICULUM VITAE

LYNN F. WOODARD



Department of Veterinary Sciences
University of Wyoming
Laramie, WY 82071
(307) 742-6638

BIRTHDATE: 6/12/46
BIRTHPLACE: Hugo, CO
SOC. SEC. NO: 517-60-5515
SPOUSE: Nancy, 12/28/53
CHILDREN: Ryan, 11/24/71

EDUCATION AND TRAINING:

Colorado State University	1968	BS	Veterinary Science
Colorado State University	1970	DVM	Veterinary Medicine
Washington State University	1978	PhD	Veterinary Microbiology
American College of Veterinary Microbiologists	1981		Board Certification

EMPLOYMENT:

1986-Present	Head and Professor, Department of Veterinary Sciences; Director, Wyoming State Veterinary Laboratory; University of Wyoming, Laramie, WY
1981-1986	Associate Professor, Department of Veterinary Science, University of Idaho, Moscow, ID
1978-1981	Assistant Professor, Department of Veterinary Science, University of Idaho, Moscow, ID
1975-1978	Postdoctoral Fellow, WOI Regional Program in Veterinary Medical Education, Washington State University, Pullman, WA
1973-1975	Self-employed, feeder cattle-farming operation, Flemingsburg, KY
1972-1973	Feedlot general manager and resident veterinarian, Greeley, CO
1970-1972	Self-employed, general veterinary practice, Glasgow, MT

PROFESSIONAL ORGANIZATIONS:

American Veterinary Medical Association
American Association of Veterinary Laboratory Diagnosticians
American College of Veterinary Microbiologists
Wyoming Veterinary Medical Association
Southeastern Wyoming Veterinary Medical Association
Gamma Sigma Delta

PUBLICATIONS:

Woodard, L.F.: Cell-Mediated Immune Responses of Cattle Following Inoculation with Tuberculo proteins Associated with a Mycobacterial Immunopotentiating Glycolipid. PhD Thesis, 1978.

Woodard, L.F., Renshaw, H.W., and Burger, D.: Cell-Mediated Immunity in Neonatal Calves: Delayed-Type Hypersensitivity and Lymphocyte Blastogenesis Following Immunization with a Mycobacterial Immunopotentiating Glycolipid and Tuberculo proteins of Mycobacterium bovis. Am J Vet Res, 39:579-584, 1978.

Woodard, L.F., Renshaw, H.W., Burger, D., McCain, C.S., and Wilson, R.B.: Cell-Mediated Immune Responses of Neonatal Calves and Adult Cattle Following Inoculation with PPD of Mycobacterium bovis Associated with a Mycobacterial Immunopotentiating Glycolipid and Oil Droplets. Am J Vet Res, 40:636-644, 1979.

Woodard, L.F., Renshaw, H.W., and Burger, D.: Onset, Kinetics, and Duration of In Vivo and In Vitro Delayed Hypersensitivity Responses of Neonatal Calves Sensitized with Mycobacterial Components. Vet Microbiol, 4:169-176, 1979.

Woodard, L.F., and Litteneker, N.: Allergenic Activity and Biochemical Analysis of Three Soluble Antigen Preparations from Brucella abortus, Strain 45/20. Am J Vet Res, 41:114-116, 1980.

Woodard, L.F., Eckblad, W.P., Olson, D.P., Bull, R.C., and Everson, D.O.: Effects of Maternal Protein-Calorie Malnutrition on Lymphoblastogenic Response of Bovine Neonates Subjected to Cold Stress. Am J Vet Res, 41:561-563, 1980.

Woodard, L.F., Eckblad, W.P., Olson, D.P., Bull, R.C., and Everson, D.O.: Serum Complement Activity of Protein-Energy Malnourished Beef Cows. Am J Vet Res, 41:1546-1548, 1980.

Woodard, L.F., Eckblad, W.P., Olson, D.P., Bull, R.C., and Everson, D.O.: Effects of Maternal Protein-Energy Malnutrition and Cold Stress on Neutrophil Function of Bovine Neonates. Am J Vet Res, 41:1048-1051, 1980.

Woodard, L.F., Eckblad, W.P., Olson, D.P., Bull, R.C., and Everson, D.O.: Hemolytic Complement Levels of Neonatal Calves Delivered from Protein-Energy Malnourished Dams and Subjected to Cold Stress. Cornell Vet, 70:266-271, 1980.

Woodard, L.F., Toone, N.M., and McLaughlin, C.A.: Immunogenic Properties of Soluble Antigens or Whole Cells of Brucella abortus Strain 45/20 Associated with Immunoadjuvants. I. Soluble Antigens. Can J Comp Med, 44:453-455, 1980.

- Woodard, L.F., Toone, N.M., and McLaughlin, C.A.: Immunogenic Properties of Soluble Antigens or Whole Cells of Brucella abortus Strain 45/20 Associated with Immunoadjuvants. II. Whole Cells. *Can J Comp Med*, 44:456-458, 1980.
- Woodard, L.F., Toone, N.M., and McLaughlin, C.A.: Comparison of Muramyl Dipeptide, Trehalose Dimycolate and Dimethyl Dioctadecyl Ammonium Bromide as Adjuvants in Brucella abortus 45/20 Vaccines. *Infect Immun*, 30:409-412, 1980.
- Olson, D.P., Ward, A.C.S., Woodard, L.F., and Bull, R.C.: Antibody Response of Protein Restricted Heifers with Escherichia coli and Passive Transfer to their Progeny. *Brit Vet J*, 36:590-596, 1981.
- Woodard, L.F., Eckblad, W.P., and Olson, D.P.: Antibody and Delayed Hypersensitivity Responses of Neonatal Calves Delivered from Protein-Restricted Dams. *Am J Vet Res*, 42:220-222, 1981.
- Olson, D.P., Woodard, L.F., Bull, R.C., and Everson, D.O.: Immunoglobulin Levels in Serum and Colostral Whey of Protein-Metabolizable Energy Restricted Beef Cows. *Res Vet Sci*, 30:49-52, 1981.
- Olson, D.P., Bull, R.C., Kelley, K.W., Ritter, R.C., and Woodard, L.F.: Effects of Maternal Nutritional Restriction and Cold Stress on Young Calves: Clinical Condition, Behavioral Reaction and Lesions. *Am J Vet Res*, 42:758-763, 1981.
- Olson, D.P., Bull, R.C., Woodard, L.F., and Kelley, K.W.: Effects of Maternal Nutritional Restriction and Cold Stress on Young Calves: Absorption of Colostral Immunoglobulins. *Am J Vet Res*, 42:876-880, 1981.
- Woodard, L.F.: Do We Need Another Brucellosis Vaccine? *Modern Vet Practice*, 62:857-859, 1981.
- Woodard, L.F., Toone, N.M., and Jasman, R.L.: Brucella abortus Vaccines: Comparison of Protection Provided by Immunopotentiated 45/20 Bacterins and Live Strain 19 in Guinea Pigs. *Am J Vet Res*, 42:1959-1962, 1981.
- Eckblad, W.P., Woodard, L.F., and Lang, B.Z.: Scanning Electron Microscopy of In Vitro Serum-Mediated Destruction of Juvenile Fasciola hepatica. *J Parasitol*, 67:784-789, 1981.
- Woodard, L.F., Jasman, R.L., Farrington, D.O., and Jensen, K.E.: Enhanced Antibody-Dependent Bactericidal Activity of Neutrophils from Calves Treated with a Lipid Amine Immunopotentiator. *Am J Vet Res*, 44:389-394, 1983.
- Woodard, L.F., and Jasman, R.L.: Comparative Efficacy of an Experimental S45/20 Bacterin and Reduced-Dose Live S19 Vaccine Against Bovine Brucellosis. *Am J Vet Res*, 44:907-910, 1983.
- Renshaw, H.W., Gessner, J.W., Woodard, L.F., and Everson, D.O.: Delayed-Type Skin Hypersensitivity and in vitro Lymphocyte Immunostimulation Responses of Swine Following Inoculation with Mycobacterium avium Cell Walls and a Mycobacterial Immunopotentiating Glycolipid. *Vet Microbiol*, 8:288-292, 1983.
- Eckblad, W.P., Stiller, D., Woodard, L.F., and Kuttler, K.L.: Immune Responses of Calves Antigenically Stimulated and Challenged with Anaplasma marginale during Tick Infestation or Treatment with Dexamethasone. *Am J Vet Res*, 45:431-436, 1984.

- Eckblad, W.P., Stiller, D., Woodard, L.F., and Kuttler, K.L.: Effect of Dexamethasone on Babesiosis in Hamsters. *Am J Vet Res*, 45:1880-1882, 1984.
- Woodard, L.F., and Jasman, R.L.: Stable Oil-in-Water Emulsions: Preparation and Use as Vaccine Vehicles for Lipophilic Adjuvants. *Vaccine*, 3:137-144, 1985.
- Workman, T., Shen, D., Woodard, L. and Yilma, T.: An Enzyme-linked Immunosorbent Assay for the Detection of Bovine Antibodies to Vesicular Stomatitis Virus. *Am J Vet Res*, 47:1507-1512, 1986.
- Talcott, P.A., Koller, L.D., Woodard, L.F., and Whitbeck, G.A.: Opposing Effects of the Interferon Inducer, Avidine: Enhancement or Suppression of Tumor Growth Depending on Treatment Regime. *Intern J Immunopharm*, 8:553-559, 1986.
- Woodard, L.F.: Adjuvant Activity of Water-Insoluble Surfactants. *Lab. Anim. Sci.*, 39:222-225, 1989.
- Woodard, L.F.: Surface Chemistry and Classification of Vaccine Adjuvants and Vehicles. In Bacterial Vaccines, Vol 13 of Advances in Biotechnological Processes. Wiley-Liss, NY, p. 281-306, 1990.
- Eng, T.R., Fishbein, D.B. and the National Study Group on Rabies: Epidemiologic Factors, Clinical Findings, and Vaccination Status of Rabies in Cats and Dogs in the United States in 1988. *J Am Vet Med Assoc*, 197:201-209, 1990.
- Mills, K.W., Johnson, J.L., Jensen, R.L., Woodard, L.F., Doster, A.R.: Laboratory Findings Associated with Abomasal Ulcers/Tympany in Range Calves. *J Vet Diag Invest.*, 2:208-212, 1990.
- Woodard, L.F.: BVD Virus Associated with Outbreaks of Abortion, Stillbirths and Weak Calves. *Vet Med* 89:379-384, 1994.
- Malczewski, A., Jolley, W.R. and Woodard, L.F.: Prevalence and Epidemiology of Trichostrongylids in Wyoming Cattle with Consideration of the Inhibited Development of Ostertagia ostertagi. *Vet Parasitol* 64:285-297, 1996.
- OTHER PAPERS/ABSTRACTS/PROCEEDINGS:
- Woodard, L.F., Toone, N.M., and McLaughlin, C.A.: Immunogenic Properties of Soluble Antigens or Whole Cells of Brucella abortus 45/20 Associated with Mycobacterial Adjuvants. In *Proc US Anim Health Assoc*, 83rd Ann Meeting, 1979, p. 152.
- Enhanced Antibody-Dependent Bactericidal Activity of Neutrophils from Calves Treated with a Lipid Amine Immunopotentiator. Western States Food Animal Research Conference, Pullman, WA, March 16, 1982.
- Olson, D.P., Woodard, L.F., Eckblad, W.P., and Bull, R.C.: Leukocyte Functions and Serum Complement Levels in Cold-Stressed Calves. In *Second International Livestock Environmental Symposium, Book of Abstracts*, Iowa State University, 1982, p. 19.
- Woodard, L.F., Jasman, R.L., Farrington, D.O., and Jensen, K.E.: Enhanced Antibody-Dependent Bactericidal Activity of Neutrophils from Calves Treated with a Lipid Amine Interferon Inducer. In *Abstracts of 2nd International Conference on Immunopharmacology*, Washington, DC, *Internat J Immunopharmacol* 4:253, 1982.

- Bull, R.C., Everson, D.O., Olson, D.P., Woodard, L.F., and Anderson, L.E.: Blood Metabolite Levels in Pregnant Beef Heifers Restricted Prepartum in Protein and/or Energy. In Proceedings, Western Section, Am Soc Anim Sci 35, 1984.
- Olson, D.P., Woodard, L.F. and Bull, R.C.: Humoral Immune Responses of Nutritionally Restricted Pregnant Heifers and Their Cold-Stressed Progeny to Immunization. Abstract, 10th Intern Congress Biometeorol, Tokyo, 1984, p. 239.
- Woodard, L.F.: Problems Associated with BVD and Practical Means of Control. In Proceedings, The Range Beef Cow Symposium X, pp. 24-27, 1987.
- Hinds, F.C., Woodard, L.F. (eds): Proceedings of Abomasitis/Abomasal Ulcers of Young Ruminants, 1987.
- Woodard, L.F. and Mills, K.: Wyoming State Veterinary Laboratory. Cow Country, April, 1989, p. 15.
- Mills, K.W., Johnson, J.L., Jensen, R.L., Woodard, L.F.: Pathogenesis and Etiology of Abomasal Ulcers/Tympany in Range Calves. Proceedings, 5th International Symposium, World Association of Veterinary Laboratory Diagnosticians, Guelph, Ontario, Canada, June, 1989.
- Woodard, L.F.: Leptospirosis. Cow Country, December 1989, p. 13.
- Woodard, L.F.: Brisket Disease. Cow Country, November, 1990, p. 10.
- Woodard, L.F.: BVD: Cleaning Up Your Herd. Cow Country, December, 1990, p. 8.
- Woodard, L.F.: Trichomoniasis Update. Cow Country, January, 1991, p. 12-13.
- Woodard, L.F., Mills, K.W., Haven, T.R. Cavender, J.L., O'Toole, D.: Agents Associated with Bovine Abortions, Stillbirths and Weak Calves in Wyoming, 1988-1990. Abstracts. 12th Annual Food Animal Disease Research Conference, Laramie, WY, April, 1991.
- Woodard, L.F.: Selection and Timing of BVD Vaccines. Cow Country, October, 1991, p. 16-17.
- Woodard, L.F.: Was That 7-Way or No-Way? Cow Country, November, 1991, p. 10-11.
- Woodard, L.F.: Wyoming State Veterinary Laboratory: An Overview. Wyoming Quarterly Update. 10:47-48, 1991.
- Woodard, L.F.: Polio in Cattle and Sheep. Cow Country, January, 1992, p. 18.
- Woodard, L.F.: Disease Problems Associated with Common Grazing. Cow Country, April 1992, p. 9.
- Woodard, L.F.: Anaplasmosis in Cattle. Cow Country, June, 1992, p. 15.
- Woodard, L.F.: Abortions in Cattle. Cow Country, January, 1994, p.6
- Woodard, L.F. and Hixon, D.: Weaning Percentage: How Do You Compare? Cow Country, February, 1994, p. 8-9.
- Woodard, L.F.: Buying Disease Problems. Cow Country, March, 1994, p. 20 & 26.

Woodard, L., Armstrong, D. Rethorst, D. and Boland, E.W.: Increased Weight Gains in Calves Following Use of a New Low-Dose vs. Standard Dose Clostridial Vaccine. Abstract, 15th Annual Western Food Animal Disease Research Conference, Lincoln, NB, March, 1994.

Woodard, L.: Malnutrition and Starvation: Economics and Ethics. Cow Country, July-August, 1994, p. 8.

Woodard, L.: Low Cost Cattle Handling Facilities. Cow Country, September, 1994, p. 14-15.

Woodard, L.: Getting Rich at Ranching. Cow Country, February, 1995, p. 7.

Woodard, L.: Essentials of Animal Disease Prevention. Western Beef Producer.
Part 1, 3: 8-9, February #2, 1995.
Part 2, 3: 8-9, 42, March #1, 1995.
Part 3, 3: 30-31, March #2, 1995.
Part 4, 3: 8-9, 66, April, 1995

Woodard, L.: 10 Myths in Livestock Agriculture. Western Beef Producer, 4:38-39, 72, February 1996.

Woodard, L.: Managing Herd Health Costs in Today's Market. Western Livestock Journal, Western Beef Producer, Cow Country, Fence Post and other publications, 1996.

Woodard, L. and Van Campen, H.: Bovine Enemy #1. Western Beef Producer section of Farmer-Stockman Magazines (multiple), 1997.

Woodard, L.F.: Annual Reports - Wyoming State Veterinary Laboratory. 1985-1997.

TEACHING:

Patb 4110 Diseases of Food Animals and Horses, 3 cr., team taught, 1986-97, University of Wyoming
MedSci 521 Natural History of Infectious Disease (bacteriology and virology sections), 5 cr., team taught to WAMI medical students, 1980-85, UI-WSU
VS 501 Seminar, 1 cr., 1980-85, University of Idaho
VS 504 Special Topics, Var. cr., 1979-85, University of Idaho
VS WS 432 Veterinary Bacteriology, 4 cr., 1976-84, Washington State University, numerous lectures and laboratories
VS 474 Animal Diseases, 3 cr., spring 1976, University of Idaho
VS 499 Herd Health Management, 1 cr., spring 1976, University of Idaho

PRESENTATIONS:

Cell-Mediated Immunity in Neonatal Calves. Colorado State University, July 31, 1978.

Delayed Hypersensitivity Responses of Newborn Calves Inoculated with Mycobacterial Components. Montana State University, September 18, 1978.

Cellular Immunity in Neonatal Calves Following Sensitization with Tuberculoproteins and a Mycobacterial Glycolipid. University of Idaho, September 27, 1978.

Delayed Hypersensitivity Responses of Newborn Calves Inoculated with Mycobacterial Components. University of California, Davis, March 13, 1979.

Effects of Malnutrition on Bovine Immunity. Texas A & M University, Amarillo and College Station, August 27-29, 1979.

Immunogenic Properties of Soluble Antigens or Whole Cells of Brucella abortus Strain 45/20 Associated with Mycobacterial Adjuvants. U.S. Animal Health Assoc., San Diego, CA, October 29, 1979.

New Immunoadjuvants for Use in Vaccines. University of Idaho, February 12, 1980.

Progress of Development of New Brucellosis Vaccines. Idaho Veterinary Medical Association, McCall, ID, July 30, 1980.

New Brucellosis Vaccines. Idaho Cattle Feeders, Stanley, ID, August 11, 1980.

New Killed Brucellosis Vaccines. U.S. Animal Health Assoc., Louisville, KY, November 2, 1980.

Experiences with new Adjuvanted Brucella abortus 45/20 Bacterins. Brucellosis Research Conference, Chicago, IL, November 9, 1980.

Brucellosis Vaccine Research. Idaho Cattlemen's Association, Boise, ID, November 22, 1980.

Cattle Trials with a New 45/20 Bacterin. Brucellosis Research Conference, Chicago, IL, November 7, 1981.

Pepping Up Your Peptides -- Adjuvants: A Key to the Success of Genetically Engineered Vaccines. Molecular Genetics, Inc., Minnetonka, MN, March 28, 1983.

Vaccine Adjuvants and Carriers. Norden Laboratories, Inc., Lincoln, NE, December 7, 1983.

Immunomodulators: A Review. Pfizer, Inc., Ames, IA, December 8, 1983.

Development of Vaccine Adjuvants. Montana State University, July 22, 1985.

New Vaccine Adjuvants and Vehicles. University of Wyoming, January 14, 1986.

Surfactants and Surface Formation in Vaccine Adjuvants and Vehicles. 38th Annual Meeting, American Association for Laboratory Animal Science, November 10, 1987.

Snovet at the Wyoming State Veterinary Laboratory. First Symposium on Veterinary Laboratory Information Systems, Ft. Collins, CO, August 10, 1991.

Increased Weight Gains in Calves Following Use of a New Low-Dose vs. Standard Dose Clostridial Vaccine. 15th Annual Western Food Animal Disease Research Conference, March, 1994.

BVD Virus: Consequences of In-utero Infections, Diagnostic and Preventive Measures. Montana Veterinary Medical Association, Bozeman, MT, January 21, 1995.

Various Animal Health Topics. Presented to Wyoming Stockgrowers, Wyoming Woolgrowers, Wyoming Veterinary Medical Association, Range Beef Cow Symposium, Wyoming-Colorado Farm Flock Days, County Beef Seminars, etc. Over 65 presentations, 1986-1997.

WORKSHOPS:

Cancer: Perspective for Control. Invited participant. Beijing, China, August 18-21, 1985.

AVMA Management Workshop. Invited participant, Chicago, December 11-12, 1988.

STUDENT ADVISING:

Pre-veterinary students

Major Professor for following graduate students:

Yung-Fu Chang - MS

William P. Eckblad - PhD

Lynn Alderson-Smith - MS

Committee Member for eight PhD and 25 MS graduate students

COMMITTEES:

University Curriculum Committee, University of Idaho

Departmental Building and Capital Outlay Committee, UI

W-102 Technical Committee, USDA

College of Agriculture Facilities Committee, University of Idaho

University Biohazard Safety Committee, University of Idaho

Departmental Graduate Committee, University of Idaho

College of Agriculture Agriculture Today/Tomorrow Committee, University of Idaho

Research Committee - IVMA Western States Veterinary Conference

Search Committee - Head and Director, WOI-Veterinary Science, University of Idaho

WRCC-46 Advisory Committee

Search Committee - Head, Department of Animal Science University of Wyoming

Governor's Task Force on Selenium in Wyoming

ABADRL-USDA Review Team

Wyoming IRM Executive Committee and State IRM Committee

AAVLD Accreditation Committee/Review Team member

PATENTS:

Brucellosis Vaccine for Cattle Containing Mycolate Esters of Trehalose, U.S. Pat. # 4,340,588, July 20, 1982.

GRANTS AND CONTRACTS:

Pacific Northwest Regional Commission - \$107,400 - Studies of Two New Brucellosis Vaccines. Principal Investigator. 1979-81.

USDA - \$96,000 - Control of Fasciola hepatica in Cattle by Vaccination and/or Chemotherapeutic Methods. Co-Principal Investigator. 1979-81.

NIH - \$300,000 - Biomedical Research and Development Grant. Co-Investigator. 1980-83.

USDA-CSRS - \$32,500 - Neonatal Calf Research Project. Co-Principal Investigator. 1978-80.

USDA - \$750 - Brucellosis in Elk. Co-Principal Investigator. 1979.

USDA - \$1,000 - New Brucellosis Vaccines. Principal Investigator. 1979.

USDA - \$10,000 - Brucella abortus Vaccines. Principal Investigator. 1980.

USDA - \$20,000 - Experimental Vaccines for the Control of Footrot in Sheep and Cattle. Co-Principal Investigator. 1980-81.

USDA - \$9,900 - Adjuvant Activities of CP-20,961, a New Lipid Amine. Principal Investigator. 1980-81.

Idaho Beef Council - \$16,200 - Development of a Killed Brucellosis Vaccine. Principal Investigator. 1981.

USDA - \$7,000 - Effect of CP-20,961 Immunostimulant on Bovine Respiratory Disease. Principal Investigator. 1981-82.

USDA - \$7,000 - Design of Vaccine Adjuvants and Emulsions. Principal Investigator. 1982-83.

USDA - \$9,990 - Effect of Vaccinating Ovine Carriers of Footrot with a Piliated, Adjuvanted Multivalent Whole Cell Bacterin. Co-Principal Investigator. 1983-84.

USDA - \$6,150 - Assessment of Interleukin-1 as the Mediator of In Vivo Adjuvant Activity. principal Investigator. 1983-84.

Norden Laboratories - \$5,000 - Gift to vaccine research. 1984.

Idaho Beef Council - \$19,200 - Synthetic Vaccine for Vesicular Stomatitis in Cattle. Principal Investigator. 1984-1985.

United Dairymen of Idaho - \$4,980 - Detection of Salmonella dublin Carriers in Dairy Cattle. Co-Principal Investigator. 1984-1985.

USDA - \$12,000 - Role of Ia Glycoprotein Expression in Vaccine Adjuvant Activity. Principal Investigator. 1984-1985.

USDA - \$59,000 - Subunit and Synthetic Peptide Vaccines and Rapid Diagnostic Kits for Vesicular Diseases. Co-Investigator. 1985-1986.

USDA - \$5500 - Vesicle Vaccine Adjuvants. Principal Investigator. 1985-1986.

USDA - \$5500 - ELISA and Fish Kidney Disease. Co-Investigator. 1985-1986.

USDA - \$2500 - Pregnancy Protein Receptor. Co-Investigator. 1985-1986.

USDA - \$5350 - Etiology and Pathogenesis of Abomasitis Complex in Range Calves. Co-Principal Investigator. 1989.

WAES - \$25,000. Effectiveness and Profitability of Management Practices Designed to Eliminate Bovine Viral Diarrhea-Related Herd Health Problems in Wyoming Cow-Calf Herds. Co-Investigator. 1994-1995.

MISCELLANEOUS:

Board of Scientific Reviewers. American Journal of Veterinary Research, 1984-1987.

Manuscripts reviewed for Journal of Animal Science, Journal of the American Veterinary Medical Association and Journal of Wildlife Diseases.

WICHE - Wyoming Delegate, Regional Advisory Council on Veterinary Medicine. 1989-1994.

CONSULTANT:

Pfizer, Inc., Terre Haute, IN
Norden, Inc., Division of Smith, Kline & Beckman, Lincoln NB
Amgen, Thousand Oaks, CA
Miles Animal Health, Shawnee, KS
Chiron, Emeryville, CA

LICENSES:

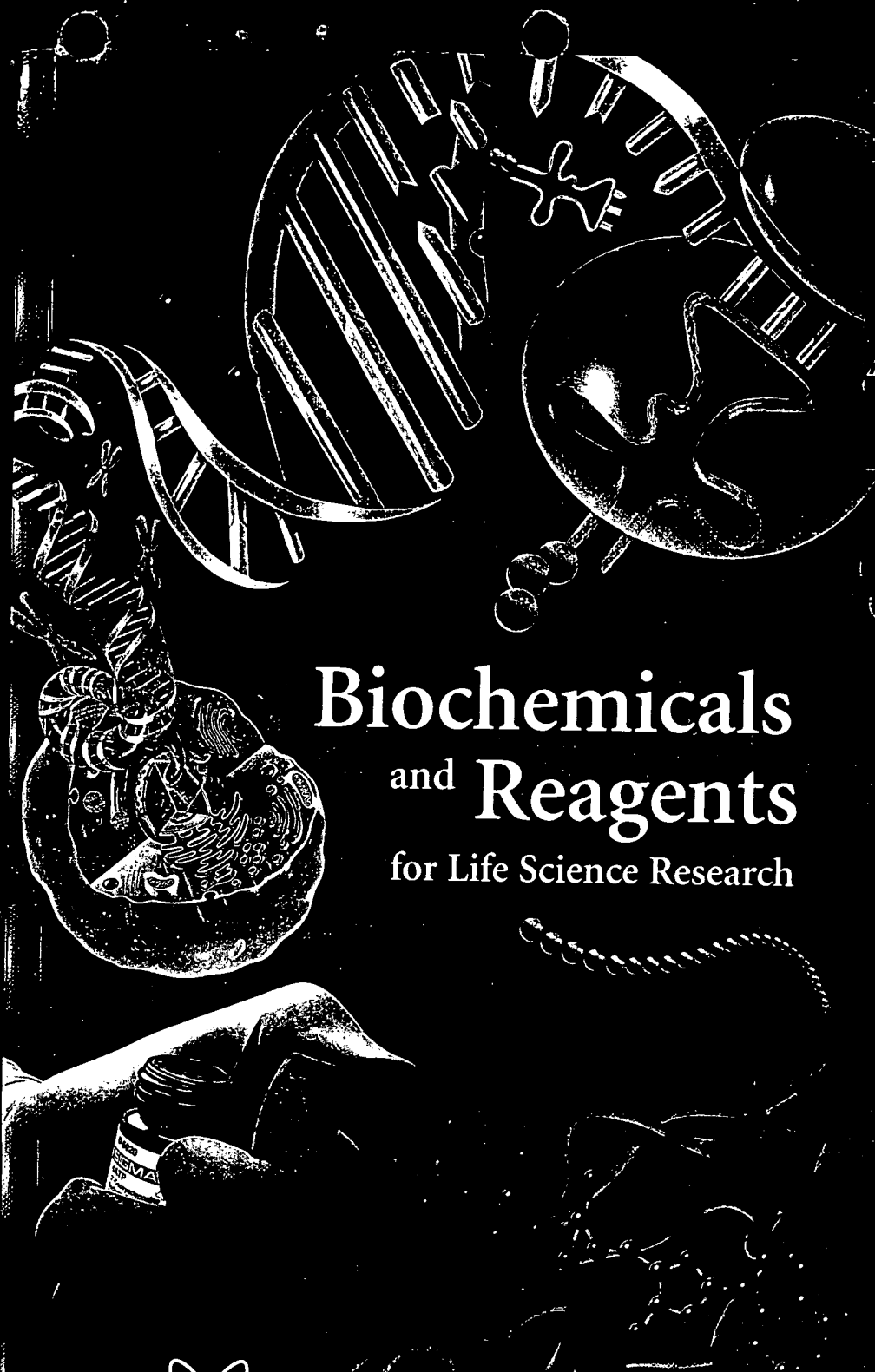
Colorado #2229 (inactive)
Montana #587 (inactive)
North Dakota #637 (inactive)
Wyoming #936 (active)

HONORS AND AWARDS:

Outstanding Young Men of America, 1981
Diplomate - American College of Veterinary Microbiologists, 1981
Outstanding Alumni Award - College of Veterinary Medicine, Washington State University, 1988.

OFFICES HELD:

President - Southeast Wyoming Veterinary Medical Association, 1987
Secretary - Southeast Wyoming Veterinary Medical Association, 1988
Secretary - WRCC-46 Research Coordinating Committee, 1988
President Elect - Wyoming Veterinary Medical Association, 1988-89
President - Wyoming Veterinary Medical Association, 1989-90
Executive Board - Wyoming Veterinary Medical Association, 1988-1991



Biochemicals and Reagents for Life Science Research

ALPHABETICAL
LIST

BIOACTIVE
PEPTIDES

IMMUNO-
CHEMICALS

MOLECULAR
BIOLOGY

RBI,
NEUROSCIENCE,
SIGNAL
TRANSDUCTION

TISSUE
CULTURE

OTHER
PRODUCT
GROUPS/USP

EQUIPMENT,
BOOKS AND
SUPPLIES

DIAGNOSTIC
KITS AND
REAGENTS

PRODUCT
INDEX



SIGMA®

(Continuation of)
POLYOXYETHYLENE ETHERS

8 Lauryl Ether (C ₁₂ E ₈) [3055-98-9] C ₂₈ H ₅₈ O ₉ FW 538.8 R: 36/37/38-41 S: 26-36	1 g 5 g 10 x 1 g	21.65 108.25 216.50
8 Myristyl Ether (C ₁₄ E ₈) [27847-86-5] C ₃₀ H ₆₂ O ₉ FW 566.8 R: 21-36/37/38 S: 26-36	1 g 5 g 10 x 1 g	21.65 108.25 216.50
8 Cetyl Ether (C ₁₆ E ₈) [5698-39-5] C ₃₂ H ₆₆ O ₉ FW 594.9	1 g 5 g 10 x 1 g	21.65 108.25 216.50
8 Stearyl Ether (C ₁₈ E ₈) [13149-87-6] C ₃₄ H ₇₀ O ₉ FW 622.9 R: 36 S: 26-36	1 g 5 g 10 x 1 g	21.65 108.25 216.50
9 Lauryl Ether (C ₁₂ E ₉ ; Polidocanol) [3055-99-0]	10 g 50 g 100 g	9.10 45.50 91.00
10 Lauryl Ether (C ₁₂ E ₁₀) [6540-99-4]	100 g 500 g 1 kg	9.10 45.50 91.00
10 Tridecyl Ether (C ₁₃ E ₁₀) [24938-91-8]	100 g 500 g 1 kg	9.10 45.50 91.00
10 Cetyl Ether (Brij 56) (C ₁₆ E ₁₀) [9004-95-9]	100 g 500 g 1 kg	9.10 45.50 91.00
10 Stearyl Ether (Brij 76) (C ₁₈ E ₁₀) [9005-00-9]	100 g 500 g 1 kg	9.10 45.50 91.00
10 Oleyl Ether (Brij 97) (C ₁₈ E ₁₀) [9004-98-2]	100 g 500 g 1 kg	9.10 45.50 91.00
20 Cetyl Ether (Brij 58) (C ₁₆ E ₂₀) [9004-95-9]	100 g 500 g 1 kg	9.10 45.50 91.00
20 Isohexadecyl Ether (Arlasolve® 200) (C ₁₆ E ₂₀) [69364-63-2]	100 g 500 g 1 kg	9.10 45.50 91.00
20 Stearyl Ether (Brij 78) (C ₁₈ E ₂₀) [9005-00-9]	100 g 500 g 1 kg	9.10 45.50 91.00
20 Oleyl Ether (Brij 98; Brij 99) (C ₁₈ E ₂₀) [9004-98-2]	100 g 500 g 1 kg	9.10 45.50 91.00
21 Stearyl Ether (Brij 721) (C ₁₈ E ₂₁) [9005-00-9]	100 g 500 g 1 kg	9.10 45.50 91.00
23 Lauryl Ether (Brij 35) (C ₁₂ E ₂₃) Suitable for use in Stein-Moore chromatography. Ref.: Moore, S. and Stein, W.H., J. Biol. Chem., 211, 893 (1954). See also: Electrophoresis Reagents Page 201 Brij 35 Solution Page 189 [9002-92-0]	100 g 500 g 1 kg 6 x 100 g 5 kg	9.10 45.50 91.00 455.00 4550.00

(Continuation of)
POLYOXYETHYLENE ETHERS

100 Stearyl Ether (Brij 700) (C ₁₈ E ₁₀₀) [9005-00-9]	250 g	9.80
W-1 May be of use as a solubilizing agent for a variety of membrane bound enzymes or in applications that call for lubrol PX and WX.	10 g 100 g 500 g 1 kg	8.00 80.00 400.00 800.00
POLYOXYETHYLENE 25 140 (PROPYLENE GLYCOL STEARATE [37231-60-0]	250 g	12.25
POLYOXYETHYLENESORBITAN		
Monolaurate (Span 20) 70% solution in water Fatty acid composition: Lauric acid approx. 50%; balance primarily myristic and palmitic acids. [9005-64-5]	250 g	8.10
Monolaurate (Tween 20) SigmaUltra Residue on ignition: <0.1% Chloride (Cl): <0.05% Sulfate (SO ₄): <0.05% Al: <0.0005% Ca: <0.0005% Cu: <0.0005% Fe: <0.0005% K: <0.005% [9005-64-5]	100 ml 500 ml	16.30 21.65
Monolaurate, Low-peroxide; Low-carbonyls (Tween 20R) Purified to remove peroxides and aldehydes Syrup Peroxides: ≤0.5 μmole/g Carbonyls: ≤1.0 μmole/g Water: ≤3% Contains BHT as an antioxidant [9005-64-5]	10 ml 5 x 10 ml 100 ml	43.50 173.90 309.80
Monolaurate (Tween 20) Syrup Fatty acid composition: Lauric acid approx. 50%; balance primarily myristic, palmitic, and stearic acids. See also: Molecular Biology Products Page 1628 and Tissue Culture Media and Reagents Page 1807 [9005-64-5]	100 ml 500 ml 1 gal 6 x 500 ml	11.50 16.20 51.35 77.55
Monolaurate, Low-peroxide; Low-carbonyls (Tween 20R) Preservative Free [9005-64-5]	10 ml 5 x 10 ml 100 ml	41.80 167.20 297.85
Monolaurate (Tween 21) Fatty acid composition: Lauric acid approx. 50%; balance primarily myristic and palmitic acids. [9005-64-5]	250 g	9.80

(Continued)

(Continuation of)
POLYOXYETHYLENESORBITAN

P 8074 Monooleate (Tween 80) SigmaUltra Residue on ignition: <0.1% Chloride (Cl): <0.05% Sulfate (SO ₄): <0.05% Al: <0.0005% Ca: <0.0005% Cu: <0.0005% Fe: <0.0005% K: <0.005% [9005-65-6] R: 40 S: 36	100 ml 500 ml	16.30 21.65
P 1754 Monooleate (Tween 80) Syrup Fatty acid composition: Oleic acid approx. 70%; balance primarily linoleic, palmitic, and stearic acids. See also: Molecular Biology Products Page 1628 Tissue Culture Media and Reagents Page 1807 and Page 1834 [9005-65-6] R: 40 S: 36	100 ml 500 ml 1 gal 6 x 500 ml	11.50 16.20 51.35 77.55
P 8192 Monooleate, Low Peroxide (Tween 80R) 10% Solution [9005-65-6] R: 40 S: 36	5 x 10 ml	52.00
P 6474 Monooleate, Low Peroxide (Tween 80R) Preservative Free [9005-65-6] R: 40 S: 36	10 ml 5 x 10 ml 100 ml	41.80 167.20 297.85
P 6349 Monooleate, Low Peroxide (Tween 80R) Preservative added [9005-65-6] R: 40 S: 36	10 ml 5 x 10 ml 100 ml	41.80 167.20 297.85
P 0343 Monooleate, Low Peroxide Non-animal source 10% Solution with BHT	5 x 10 ml	50.00
P 2815 Monooleate (Tween 81) Fatty acid composition: Oleic acid approx. 70%; balance primarily palmitic and linoleic acids. [9005-65-6] R: 40 S: 36	250 g	9.05
P 1504 Monopalmitate (Tween 40) Syrup Fatty acid composition: Palmitic acid approx. 90%; balance primarily stearic acid. [9005-66-7]	100 ml 500 ml 1 gal	11.50 16.20 51.35
P 1629 Monostearate (Tween 60) Fatty acid composition: Stearic acid approx. 50%; balance primarily palmitic acid. [9005-67-8]	100 ml 500 ml 1 gal	11.50 16.20 51.35
P 3065 Monostearate (Tween 61) Fatty acid composition: Stearic acid approx. 50%; balance primarily palmitic acid. [9005-67-8]	250 g	8.80

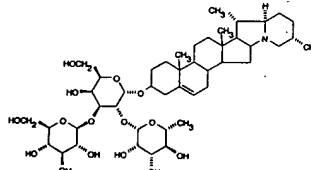
(Continued)

P

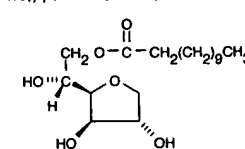
BEST AVAILABLE COPY

LIST OF COMPOUNDS

ALPHABETICAL LIST OF COMPOUNDS

PRODUCT NUMBER	US \$
SODIUM TUNGSTATE	100 g 12.35 500 g 5.71
S 0765 Dihydrate ACS Reagent Assay: 99.0 to 101.0% Insoluble matter: ≤0.01% Titratable free base: ≤0.02 meq/g Chloride (Cl): ≤0.005% Molybdenum (Mo): ≤0.001% Nitrogen compounds (as N): ≤0.001% Sulfate (SO ₄): ≤0.01% Heavy metals and iron (as Pb): ≤0.001% [10213-10-2] Na ₂ WO ₄ • 2H ₂ O FW 329.9 R: 20/21/22-36 S: 26-36	
SODIUM TUNGSTATE See: Tungstic Acid, Sodium Salt Page 1048	
SODIUM 12-TUNGSTOPHOSPHATE See: Phosphotungstic Acid, Sodium Salt Page 846	
SODIUM UNDECYLENATE See: Undecylenic Acid, Sodium Salt Page 1053	
SODIUM URATE See: Uric Acid, Sodium Salt Page 1055	
SOLANESOL	25 mg 52.30 100 mg 153.80 250 mg 306.15
S 8754 From Tobacco leaves 90+% (HPLC) This C ₂₅ isoprenoid alcohol is the most abundant lipid in tobacco leaves. It may be an important precursor of the tumorigenic polynuclear aromatic hydrocarbons of smoke. Ref.: Severson, R.F., et al., J. Chromatog., 139, 28 (1977). [13190-97-1] C ₂₅ H ₅₂ O FW 631.1	
SOLANIDINE	5 mg 21.60 10 mg 35.10 50 mg 136.10
S 7881 From Potato Approx. 98% Ref.: Ripberger, H. and Schreiber, K., The Alkaloids, XIX, 81 (1981). [80-78-4] C ₂₇ H ₄₃ NO FW 397.6 R: 23/24/25 S: 45-36/37/39-22	
α-SOLANINE	5 mg 43.35 10 mg 71.35 50 mg 235.10
S 3757 From Potato sprouts Approx. 95% A trisaccharide, consisting of glucose, galactose and rhamnose, linked to solanidine. Ref.: Bushway, R.J., Amer. Potato J., 60, 293 (1983).	
 See also: α-Chaconine Page 243 [20562-02-1] C ₄₅ H ₇₃ NO ₁₅ FW 868.1 R: 23/24/25 S: 45-36/37/39	
SOLANINE D See: Demissidine Page 329	
SOLANUM TUBEROSUM LECTIN See: Lectins Page 2095	
SOLASODINE	100 mg 29.20 250 mg 56.70 1 g 165.05
S 0888 (Solasonod-5-en-3β-ol) Approx. 99% [126-17-0] C ₂₇ H ₄₃ NO ₂ FW 413.6 R: 23/24/25 S: 45-36/37/39	

PRODUCT NUMBER	US \$
SOLKETAL	500 g 32.00
S 6635 (1,2-O-Isopropylidene-rac-glycerol; (1±)-2,2-Dimethyl-1,3-dioxolane-4-methanol) Approx. 97% d = 1.07 g/ml For possible use in lipid synthesis. Aldrich Brand. Formerly Sigma Product I 8380. [100-79-8] C ₈ H ₁₂ O ₃ FW 132.2	
SOLVENT BLUE 37	10 g 10.90 25 g 21.60
S 2229 Practical Grade [12226-74-3]	
SOLVENT BLUE 38	25 g 19.90 100 g 40.15
S 2232 Practical Grade [1328-51-4]	
SOLVENT GREEN 4 See: Fluorol Yellow 088 Page 455	
SOLVENT RED 27 See: Oil Red O Page 773	
SOMATOSTATIN and RELATED PEPTIDES See: Bioactive Peptides Page 1126 For cell culture tested Somatostatin See: Tissue Culture Media and Reagents Page 1808	
SOMATOTROPIN (STH) (Growth Hormone) Increases mass of most tissues by increasing cell number rather than cell size, mobilizes fat stores [9002-72-6]	
S 4776 From Human Pituitaries 1 vial 148.40	
S 4776 Vial contains approx. 4 I.U. h-GH, with 0.1 mg ammonium bicarbonate and 3.5 mg mannitol. Not assayed by Sigma.	
S 6848 From Porcine Pituitaries 1 vial 90.55	
S 6848 Vial contains approx. 50 I.U. Bioassay not run by Sigma.	
SOPHORA JAPONICA See: Lectins Page 2096	
SOPHOROSE	10 mg 33.60 25 mg 66.35 100 mg 220.70
S 1404 (2-O-β-D-Glucopyranosyl-β-D-Glc-[1→2]-β-D-Glc) Minimum 98% [20429-79-2] C ₁₂ H ₂₂ O ₁₁ FW 342.3	
SORBIC ACID (2,4-Hexadienoic acid)	
S 1626 Free Acid	100 g 13.35 250 g 17.15 500 g 25.95
S 1626 Minimum 99.0% See also: Tissue Culture Media and Reagents Page 1836 [110-44-1] C ₆ H ₈ O ₂ FW 112.1 R: 20/21/22 S: 26-36	
S 17420 Potassium Salt SigmaUltra	100 g 25.80 500 g 59.50
S 17420 Minimum 99% Insoluble matter: <0.1% Solubility (1 M in water, 20°C): complete, faint yellow Cl: <0.05% Mg: <0.0005% Na: <0.1% NH ₄ : <0.05% P: <0.001% Pb: <0.001% Zn: <0.0005% [590-00-1] C ₆ H ₅ O ₂ K FW 150.2 R: 36/37/38 S: 26-36	
S 1751 Potassium Salt (Minimum 99%) [590-00-1] C ₆ H ₅ O ₂ K FW 150.2 R: 36/37/38 S: 26-36	100 g 12.25 250 g 16.55 500 g 30.95

PRODUCT NUMBER	US \$
SORBITAN	
S 6635 Monoaurate (Span 20) Fatty acid composition: Lauric acid (C12:0) approx. 50%; balance primarily myristic (C14:0), palmitic (C16:0) and linolenic (C18:3) acids.	250 ml 16.15 1 liter 49.95
 [1338-39-2]	
S 6760 Monooleate (Span 80) Fatty acid composition: Oleic acid (C18:1) approx. 75%; balance primarily linoleic (C18:2), linolenic (C18:3) and palmitic (C16:0) acids.	250 ml 14.15 1 liter 45.00
S 6885 Monopalmitate (Span 40) Fatty acid composition: Palmitic acid (C16:0) approx. 90%; balance primarily stearic acid (C18:0). [26266-57-9] R: 36/37/38 S: 26-36	250 g 14.15 1 kg 45.00
S 7010 Monostearate (Span 60) Fatty acid composition: Stearic acid (C18:0) approx. 50%; balance primarily palmitic acid (C16:0). [1338-41-6]	250 g 15.10 1 kg 47.85
S 3386 Sesquileate (Arlacel 83) Fatty acid composition: Oleic acid (C18:1) approx. 70%; balance primarily palmitic acid (C16:0), stearic acid (C18:0) and linoleic acid (C18:2). [8007-43-0] R: 36/37/38 S: 26-36	250 ml 14.15 1 liter 45.00
S 7135 Trioleate (Span 85) Fatty acid composition: Oleic acid (C18:1) approx. 74%; linoleic acid (C18:2) approx. 7%; linolenic acid (C18:3) approx. 2%; palmitoleic acid (C16:1) approx. 7%; balance primarily palmitic acid (C16:0). [26266-58-0] R: 36/37/38 S: 26-36	250 ml 14.15 1 liter 45.00
S 2028 Tristearate (Span 65) Fatty acid composition: stearic acid (C18:0) approx. 50%; balance primarily palmitic acid (C16:0). [26658-19-5]	250 g 14.05 1 kg 44.55